British Society for Parasitology

52nd Annual Spring meeting and Trypanosomiasis and Leishmaniasis symposium

6th to 9th April 2014
University of Cambridge

Scientific program and abstracts

A Warm Welcome

It is our great pleasure to extend a warm welcome to all delegates to the 52nd Spring Meeting of the British Society for Parasitology and Trypanosomiasis/Leishmaniasis symposium, at the University of Cambridge. This is the first Spring Meeting at the University of Cambridge, and we hope that this is a successful, productive and enjoyable meeting for you all. Cambridge offers a near unique venue, and we also hope that delegates can enjoy the special atmosphere of the City and the Fenlands if they have the time. The meeting is based in Robinson College, a superb site for scientific meetings, with many of the larger events taking place in the West Road Concert Hall, which is part of the Faculty of Music; these venues are only five minutes apart and ten minutes from central Cambridge (see maps). Please check carefully that you are in the correct place for your sessions, and please ask our helpers if you need assistance.

Cambridge has a long history of association with Parasitology. To take just one example, Cambridge was the place where the variant surface antigen was first described, and is also the home of the Molteno Institute (founded in 1921 and now simply part of the Department of Pathology), which gave its name to many of the classic trypanosome strains as "MITat" (Molteno Institute Trypanozoon antigen type). Establishing the Molteno and the journal Parasitology were dual achievements of the very eminent George Nuttal in the early 20th Century. A brief history of the Molteno, its origins, importance and the contributions of the Institute to parasitology can be found at:
http://www.path.cam.ac.uk/~schisto/home/molteno.html

We hope that the 2014 meeting will be one to remember, and we have an exceptionally packed and varied timetable with, we hope, something for everyone. The scientific sessions cover a full range of interests for parasitologists, and also extend into special sessions beyond these core disciplines, including a session focusing on research in the Middle East and a specific evolution mini-session. We also have a lively communications/outreach program, with guest lectures and the Naked Scientists and their Radio show, to be recorded live as a podcast. With over 500 participants we are sure that new collaborations and friendships will emerge from the meeting. We also hope that the city itself will play a starring role, with a reception on Sunday evening at the Fitzwilliam Museum and the Conference Dinner and traditional ceilidh at Robinson College on the Tuesday. A Young Parasitologist Party, with some of our members playing the Blues will take place Monday evening. The now regular BSP debate, Wright medal lectures and the AGM help to round out our program.

We thank you for attending the BSP Spring meeting, and hope that you find it stimulating, enjoyable and fun.

Mark C. Field, University of Dundee
Owain Millington, University of Strathclyde

BSP 2014 Organisers

Contents

The BSP Council 2014

The BSP Council meets three or four times a year and has one or two additional teleconferences. It is made up of a board of trustees plus non-trustee members.

Board Trustees:

Professor Simon Croft: President
Professor Judith Smith: Vice-President/President elect
Professor J. Russell Stothard: Honorary General Secretary
Professor Angus Bell: Honorary Treasurer
Dr Owain Millington: Honorary Meetings Secretary
Dr Emily Adams: Honorary Communications Secretary
Professor Mark C. Field: Ordinary council member
Dr Paul Denny: Ordinary council member
Dr Catherine Merrick: Ordinary council member
Dr Damer Blake: Ordinary council member

Non-Trustees:

Emily Dawson: Student Representative
Luke Roberts: Student Representative

Finance Sub-committee:

Professor Angus Bell
Dr Paul Denny
Dr Catherine Merrick

Do you want to become more involved with the BSP?

The BSP is seeking new members to sit on council when present members rotate off after their tenure. The usual term is three years, and is an opportunity to contribute to the running of a society that seeks to promote parasitology in all its forms. The BSP council meets about four times a year, at various locations around the UK, and is primarily responsible for the smooth running of the Spring and Autumn meetings, together with outreach and other aspects. If you are interested, please talk to any of the present council members listed above or contact the BSP Secretariat at *info@bsp.uk.net*.

BSP AGM

West Road Auditorium, 10am Wednesday 9th April

A new BSP constitution, suitable for taking the Society forward for the next decade, has been drafted, and which seeks to update the way the society is run in response to new media and changes in the charitable organisation landscape. It is highly important for BSP members to have the opportunity to discuss this new constitution, and the AGM provides the major forum for this. The council would encourage you to attend and make a contribution; it is only through having your opinions heard that the Society can adapt and change in the manner you wish it to, and we hope to see you there.

Conference venues, West Road and Robinson College

We politely request that, out of respect for all speakers, debaters and other presenters that you switch your mobile/cellular phone to silent/vibrate whenever you are in an auditorium with an active session in progress.

Reception and registration

Registration for the meeting will be from 12.00 noon to 6.00 pm on Sunday 6th of April in Robinson College. The reception desk will re-open at 9:00am on Monday the 7th of April in the West Road Concert Hall (Maps pages 10-12).

Other facilities that will be made available at the reception desk will include:

- Urgent message pickup
- Conference organising committee contact details
- Internet access codes
- An information/messages board
- Tourist Information
- Access to conference rooms for project meetings

Urgent HELP!!

You need urgent help and I don't know what to do and it's an emergency: Call Julian and the BSP Secretariat on 0775 7980894.

Safety

On the continuous sounding of a fire alarm, evacuate by the nearest safe route to the assembly areas. For West Road this will be on the walkway opposite the main entrance and for Robinson in the area immediately outside of the main court. Emergency exits are clearly identified with illuminated green signs.

Please comply with all fire regulations, as well as instructions of staff at both venues. This is especially important in crowded lecture theatres – please ensure that aisles and walkways are kept clear.

In the event of an emergency, dial 1-999 from any internal phone to contact emergency services.

Identification

We would like to remind all delegates that it is important to wear name badges at all times, in order to identify yourselves to the organizers, volunteer helpers and university staff. The conference committee can be identified by name badges which display the BSP 2014 logo and student volunteers who will be wearing BSP 2014 polo-shirts. All of these people are here to help you, so please do not hesitate to ask for assistance.

Internet Facilities

Delegates can access the internet via several different wireless networks. The University of Cambridge has guest Wi-Fi passes valid for the duration of the conference *via* the Lapwing network. Users who have an Eduroam account can also use this to log in directly to the system, and will likely find this to be the most efficient option. Access codes and details of how to sign onto Lapwing can be picked up from the Conference Reception desk. If there are any problems, please contact the Conference Registration desk who can provide details of IT Services.

CONFERENCE WIRELESS INTERNET ACCESS

Robinson College

In Order to log on to the Robinson College Conference Wireless Network, please follow the instructions as below:

- Log on to Wireless Network: Rob-Conf
- Type in password: december1122
- Open a public page i.e. Google, yahoo etc.
- You will then have to click 'I Agree' to the terms and conditions
- Access Internet!

Please Note: The Internet connection may disconnect intermittently/daily so please repeat the steps above if this occurs.

Support

The College is not able to provide IT support to individual delegates. However, our IT Department will ensure that the network and system are fully functioning and whenever possible, will offer advice to resolve problems. Please note that the IT Department is only open during normal office hours. In the event of difficulties, delegates should be advised to report the problem to the Porters Lodge who will record information for attention by the IT Department at the next available opportunity.

Central Cambridge

Information for oral presentations

Oral presentations will be given in the Auditorium, Recital room and Lecture room in West Road, and in the Auditorium, Umney Lecture Hall and the Junior Common Room in Robinson College (see floor plans on pages 11 and 12).

Loading of oral presentations will take place in the lecture theatre appropriate to the session, and there is either a desktop of laptop computer provided. Speakers may use their own laptops if required, but please ensure you bring the appropriate video and power adaptors.

Please ensure that your presentation is loaded by 8:45 am at the latest if you are presenting in the morning sessions or by 1:45 pm if you are presenting in the afternoon sessions.

There will be a student volunteer in each room who will help you load your presentation and assist you if any problems arise with the A/V equipment.

The scientific program is very full and speakers are respectfully requested to keep to their time slot so that delegates who wish to move between sessions can do so. Chairs have been instructed to be strict with their timings.

Information for Poster Presentations

Posters may be put up from 4:30 pm on Sunday 6th April at the West Road Concert Hall, and the boards (6ft tall x 3ft wide) are located in the lobby area as well as some associated spaces adjoining. You will be provided with a number for your poster that will correspond to a particular poster board. Please note that the use of blue tack or drawing pins is prohibited and only the Velcro tabs provided may be used to secure your poster to the boards. These will be made available at the Conference Reception desk.

All posters must be displayed before the poster session (between 6:00 and 8:00 pm Monday), and presenters should stand with their poster during this time.

Posters must be removed by 5:30 pm on Tuesday. BSP cannot store or return posters after this time.

Student Prizes

Prizes for the best oral and poster presentations by student delegates have been generously provided by Biomed Central on behalf of the *Malaria Journal* and *Parasites and Vectors*, and by Cambridge University Press on behalf of *Parasitology*.

Eligible entrants for the student competitions are marked with and asterisk (*) throughout the program and abstracts. Please use the tear out voting slip at the back of the abstract

book to register your vote; these should be handed in to the Conference Reception desk at the end of the poster session on Monday.

Student travel awards

These are collected from the Secretariat straight after the AGM which you will need to attend. You will have to sign for it and should have some form of photo I.D. with you. Payment is by cheque, and any overseas members can request a bank transfer in place of this. Cheques have a limited life and need to be deposited soon after the meeting. Beyond a certain point we are unable to complete awards to students who loose or misplace cheques and require payment at a much later date.

Food and Refreshments

Breakfast is available in the Garden Restaurant until 9.00 am for those staying at Robinson College. This is a staff-assisted self-service meal. A full range of cooked items and cereals is available as well as continental breakfast. A self-clearing system operates in the Garden Restaurant area.

All coffee breaks will be provided in either the lobby area of West Road Concert Hall or the breakout areas of Robinson College.

Lunch will be served in the Dining Hall/Garden Room at Robinson College (Floor plan page 11).

Delegates are reminded to ensure that their name badges are easily seen by University staff serving food and refreshments.

Packed lunches on Wednesday

We will provide a number of packed lunches to see you on your way after the meeting closes. To book a packed lunch just drop a penny in the jar on the registration desk, one for each lunch required and you can collect from Robinson at the close of the meeting

In addition to the refreshments and meals provided by the conference, the Red Brick Café will be open from 9.30 am to 11.00 pm throughout the conference for purchase of coffees, other hot beverages and light bites, as well as an interesting choice of cask conditioned beers, lagers and wines as well as the full range of spirits. The Bar is run on a cash basis, and credit cards are accepted in the Bar (minimum spend £10).

There are hot and cold beverage and confectionery vending machines situated in the Bar/JCR Foyer area.

Information of delegates resident at Robinson College

Rooms are supplied with soap and towels; beds are made with duvets in covers; extra blankets may be obtained from the bedmaker on request. Rooms are serviced daily by bedmakers. Radio alarm clocks, tea and coffee making facilities and bottled mineral water are provided in all bedrooms. The supplies will be replenished by the bedmaker. Shaver socket outlets provide 115 or 230 volt supply. Electrical adapters are available from the Porters' Lodge; voltage in the UK is 240 A/C. Internet access is free of charge. All of our bedrooms have internet access via Wi-Fi or a wired network point.

On the day of departure, BEDROOMS MUST BE VACATED AND KEYS RETURNED TO THE PORTERS' LODGE BY 09.30am, to enable rooms to be serviced for incoming delegates. Keys should be returned to the Porters' Lodge and a charge of £23.50 per key will be made if this is not done.

ALL VISITORS ARE STRONGLY RECOMMENDED TO ENSURE THAT ROOM DOORS ARE LOCKED AND WINDOWS ARE CLOSED WHENEVER ROOMS ARE UNOCCUPIED.

We draw your attention to the fact that in no circumstances can the College accept responsibility for the loss of or damage to any property brought to the College including motor cars.

Smoking policy: *http://www.robinson.cam.ac.uk/conferences/smoking.php*

Safety policy: *http://www.robinson.cam.ac.uk/about/publications/robinson_safety_policy.pdf*

Access needs: *http://www.robinson.cam.ac.uk/conferences/accessibility.php*

Incoming mail will be delivered to you daily and should be addressed as follows:

BSP Spring meeting 2014,
Robinson College,
Cambridge,
CB3 9AN
UK

Outgoing stamped mail can be posted in the Porters' Lodge. Postage stamps are also on sale in the Porters Lodge.

Taxi

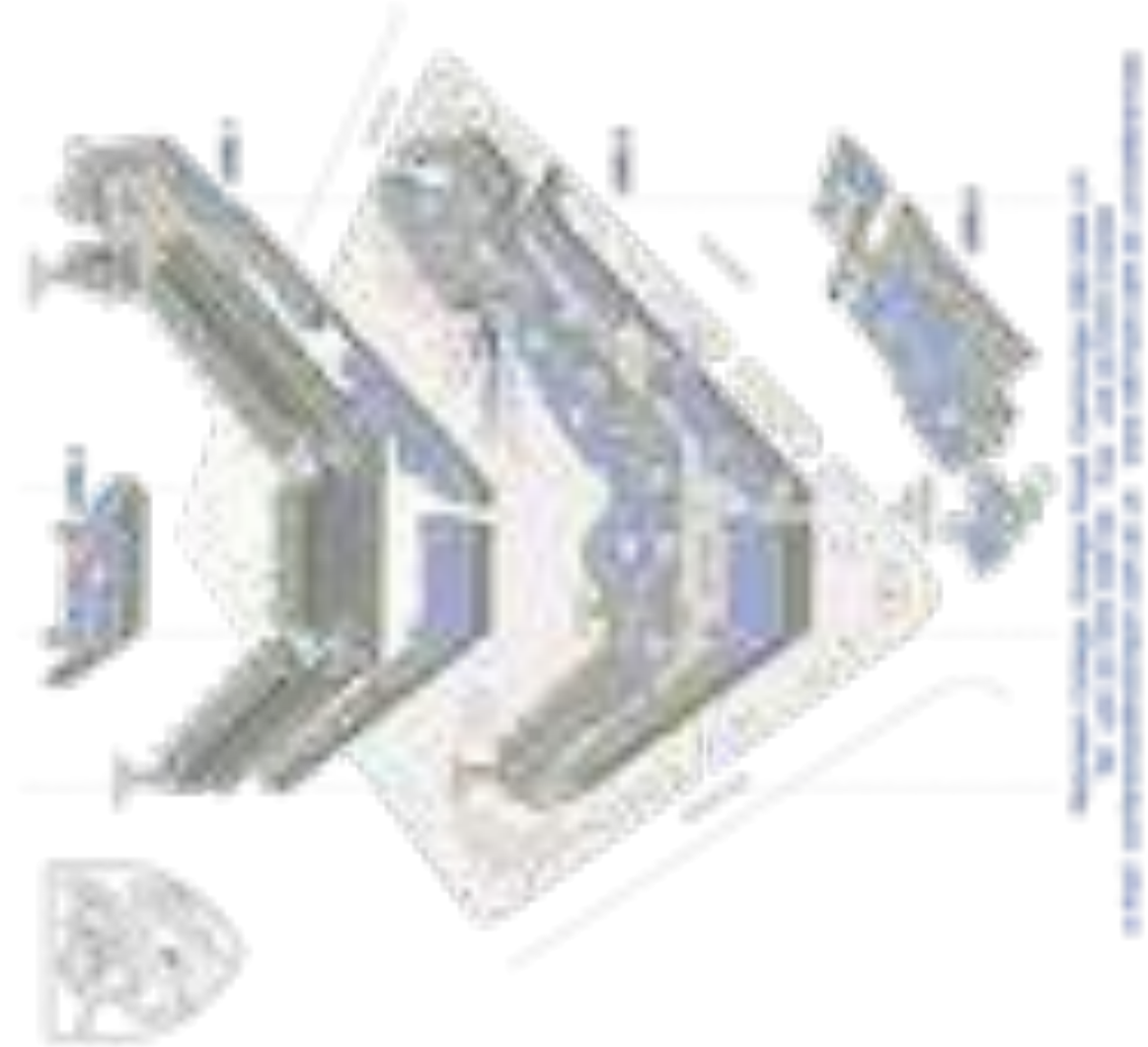
The Porters' Lodge can order taxis on request. It is helpful if delegates who require a taxi on departure give their request to the Porters' Lodge the night before leaving to ensure that sufficient taxis are available. Alternatively call Panther Taxi on 01223 715715.

Wired Network

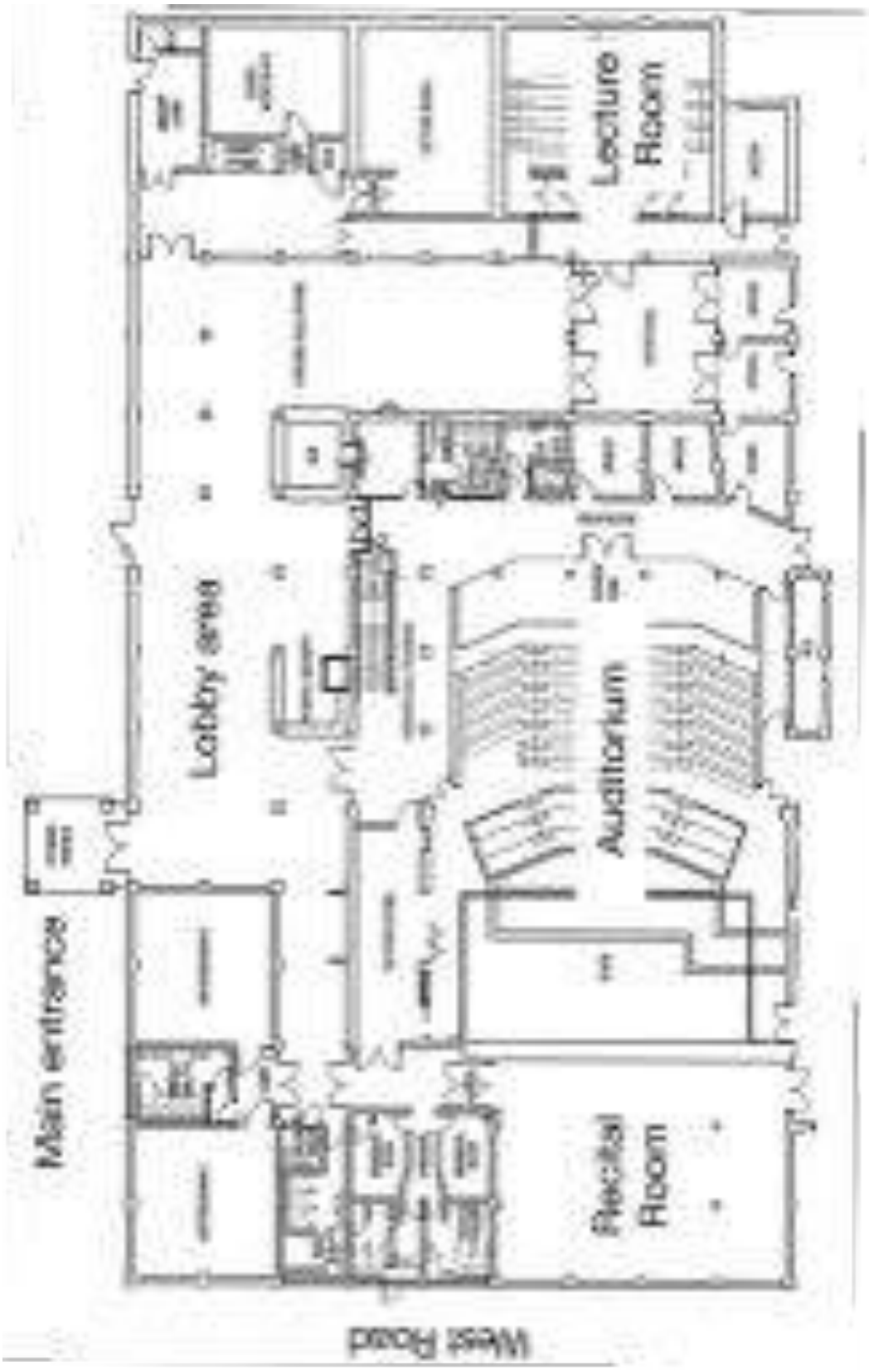
Robinson College also offers free internet connections via a high-speed local network. All meeting rooms, auditoria, and bedrooms are served with 100Mb wired connections. CAT-5 ethernet cables are available for purchase from the Porters' Lodge. Please refer to the website for further details and important information:
http://www.robinson.cam.ac.uk/conferences/facilities/internet.php

Left Baggage Facilities

On Wednesday, delegates wishing to do so may leave their luggage in ‘left luggage’ at Robinson up to 2:00 pm. Signs will indicate the room but please ask if in doubt.



Floor plan 1: Robinson College



Floor plan 2: West Road Concert Hall; public areas

Social Program and Trade Fair

Sunday 6th April: To kick off the meeting, all delegates are invited to a welcome drinks reception at the European galleries of the magnificent Fitzwilliam museum, to be held from 7:00 pm. This grand and imposing building, housing a superb collection of fine art, archeological pieces, arms, armour, ceramics and more, is based on Trumpington Street. Short welcome speeches will be given from the current BSP President and Deputy Vice Chancellor for the University of Cambridge, but ample time is available to view works by the French Impressionists, English School and more.

Monday 7th April: At 5:00 pm there will be a public lecture by David Dunne, University of Cambridge, describing his experiences as a scientist working extensively in Africa. The special challenges of co-ordinating such long distance teams will be discussed. This will be followed by our poster event, with beer and wine reception, from 6.00pm. The poster session will be held in the lobby area of the West Road Concert Hall.

The **BES Special Interest Group** on Parasite Ecology will be hosting a social event on ***Monday 7 April from 7pm*** at the Anchor Pub. Everyone attending the Ecology stream is welcome. A buffet with nibbles and drinks will be provided, first come first served. The pub is located by the river on Silver Street, opposite Queens College, 10 minutes from the venue. More info on the Group is available at:
http://www.britishecologicalsociety.org/getting-involved/special-interest-groups/parasite-and-pathogen-ecology-and-evolution/

All delegates are invited to the book launch of eParasitism, 2nd Edition at ***6pm on Monday 7th*** and to meet the authors. The launch is at the Cambridge University Press Bookshop on the Market Square, about 10 minutes walk from West Road/Robinson College. **Please email Fran Robinson at** *frobinson@cambridge.org* **if you would like to attend.**

The Young Parasitologists Party will be in the Avery pub (Regent St) from ***8:00 pm after the Poster viewing on Monday 7th***. All students will have tickets for this in their delegate packs. Come join us for the first evening of the Spring meeting, for a food buffet, some complimentary drinks and some light entertainment to get everyone in the conference spirit! Grab someone you don't recognise, get to know each other and form some teams for the **YPP quiz** in the first half of the evening at 8:30pm and who knows what fabulous prizes may await you! Afterwards, be entertained from 9:45pm by the brilliant '**Dr Snow**'**s Pumphandle band**, composed of an array of some of your favourite in-house BSP members who will play a variety of tunes as a soundtrack for continued merriment into the night. Audience participation will be encouraged and opportunities to grace the stage will arise so warm up those vocal cords and get involved! Hope to see you there!

Tuesday 8th April: Conference dinner at Robinson College. The evening commences with a welcome drinks reception at 7.00 pm. Dinner will be served at 8.00 pm. After this the room will be cleared for the Ceilidh by Teannaich. Dancing finishes at 11.30 pm.

Eating and drinking

For other places to eat and drink and catch up with friends and colleagues, your conference delegate packs will include a City Guide to help you find the many excellent pubs, bars, restaurants and shopping to keep you entertained. If you are extending your stay then additional tourist information will also be available at the Conference Reception desk. Also, visit

http://www.cambridge-pubs.co.uk and *http://www.tripadvisor.com/Restaurants-g186225-Cambridge_Cambridgeshire_England.html*

for dining suggestions and

http://www.visitcambridge.org/visitor-information/tourist-information-centre for general tourist information.

Trade fair

A trade fair, featuring several publishers and technology companies will run during most of the meeting in the West Road Auditorium lobby. Please do take a chance to visit, to meet the exhibitors and to take the opportunity to discuss your needs and ideas with them.

Timetable at a glance

We politely request that, out of respect for all speakers, debaters and other presenters that you switch your mobile/cellular phone to silent/vibrate whenever you are in an auditorium with an active session in progress.

	Sunday 6th April					
12:00 - 18:00	Registration					
19:00	**Reception Fitzwilliam Museum, Welcome address**					
	Monday 7th April					
9:00 -10.30	**Plenary lectures:** Tom Blundell, Bryan Grenfell and Keith Gull					
10:30 -11:00	**Coffee break**					
	Session A West Road Recital*	**Session B** West Road Lecture Room	**Session C** West Road Auditorium*	**Session D** Robinson Auditorium	**Session E** Robinson Umney	**Session F** Robinson JCR
11:00 - 12:30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
12:30 -14:00	**Lunch and Naked Scientists**					
14:00 -15.30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
15:30 -16:00	**Coffee break**					
16:00 -17:30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
17:30 -18:00	**The Africa Experience:** David Dunne					
18:00 -20:00	**Poster session and trade fair**					
20:00 on	**Young parasitologist party:** The Avery, Regent Street					

*Note session A & C swap locations on day 2.

	Tuesday 8th April					
09.00 -9.30	**Plenary lecture**: David Roos					
9.30 -10.30	**CA Wright medal lectures** Alex Rowe and Michael Barrett					
10:30 -11:00	**Coffee break**					
	Session A West Road Auditorium*	Session B West Road Lecture Room	Session C West Road Recital*	Session D Robinson Auditorium	Session E Robinson Umney	Session F Robinson JCR
11.00 -12.30	Tryp/leish	Apicomplexa	Evolution	Helminths	Communication	Veterinary
12:30- 14:00	**Lunch and social media**					
14.00 -15.30	Tryp/leish	Apicomplexa	Evolution	Helminths	Ecology	Careers
15:30 -16:00	**Coffee break**					
16.00 - 17:30	Tryp/leish	Apicomplexa	Mid East	Helminths	Ecology	Veterinary
15:30 -16:00	**Gala dinner: Robinson College**					
	Wednesday 9th April					
9.30 -10.30	**BSP Debate: Vector control** **BSP AGM**					
10:30 -11:00	**Coffee break**					
	Session A West Road Auditorium	**Session B** West Road Lecture Room	**Session C** West Road Recital	**Session D** Robinson Auditorium	**Session E** Robinson Umney	**Session F** Robinson JCR
11.00 -12.30	Tryp/leish	Apicomplexa	Mid East	Helminths	Ecology	Veterinary
12:30- 14:00	**Lunch and meeting close**					

*Note session A & C swap locations on day 2.

Detailed Program and Agenda

Registration - Sunday 6th April 2013

Registration: Robinson College 12:00 PM - 6:00 PM (360 mins)

Reception

Fitzwilliam Museum 7:00 PM - 9:00 PM (120 mins)

Welcome address: Prof Simon Croft, BSP President
7:45 PM - 8:00 PM (15 mins)

Registration - Monday 7th April

Registration: West Road 8:30 AM - 5:29 PM (539 mins)

Plenary sessions 1, 2 and 3 (WRA): Profs Bryan Grenfell, Keith Gull, Tom Blundell

Plenary sessions 1, Profs Bryan Grenfell 9:00 AM - 9:30 AM (30 mins)
Microparasites in space and time (Bryan Grenfell)

Plenary sessions 2, Keith Gull 9:30 AM - 10:00 AM (30 mins)
The extreme evolutionary cell biology of trypanosomes. (Keith Gull)

Plenary sessions 3, Tom Blundell 10:00 AM - 10:30 AM (30 mins)
The development of targeted anti-microbials to combat tuberculosis and the emergence of resistance (Tom Blundell)

Morning Refreshments

Coffee and Tea Break 10:30 AM - 11:00 AM (30 mins)

Session A1 (WRL) - Kinetoplastida - Genome structure and evolution - Chair: Steve Kelly, University of Oxford

A1-IS - Invited speaker Andrew jackson, University of Liiverpool 11:00 AM - 11:30 AM (30 mins)
Slimming down and suiting up: key transitions in the evolution of trypanosomatids (Andrew Jackson)

A1-O1 11:30 AM - 11:45 AM (15 mins)
Insights into 'missing biology' of Leishmania mexicana from RNA-sequencing (Eva Gluenz)

A1-O2 11:45 AM - 12:00 PM (15 mins)
Nuclear DNA Replication in Trypanosoma brucei: the beginning (Catarina A. Marques)

A1-O3 12:00 PM - 12:15 PM (15 mins)
The genome of Trypanosoma rangeli, a trypanosomatid avirulent to mammals (Edmundo Grisard)

A1-O4 12:15 PM - 12:30 PM (15 mins)
Aneuploidy in natural Leishmania populations: chromosome chaos or adaptive strategy? (Jean-Claude Dujardin)

Session B1 (WRR) - Apicomplexa - Cell Biology - Chair: Julian Rayner, Wellcome Trust Sanger Institute

B1-IS - Invited speaker Jacob Baum, Imperial College London 11:00 AM - 11:30 AM (30 mins)
Single Molecular, Structural and Biochemical insights into actin regulation in the malaria parasite (Jacob Baum)

B1-O1 11:30 AM - 10:45 AM (-45 mins)
Adhesion properties of P. falciparum infected erythrocytes that bind human brain endothelial cells (Yvonne Azasi)

B1-O2 11:45 AM - 12:00 PM (15 mins)
Exploring the membrane-microtubule interaction at the cortex of Toxoplasma gondii (Ke Hu)

B1-O3 12:00 PM - 12:15 PM (15 mins)
Processing of Plasmodium falciparum merozoite surface protein 1 is essential for blood stage parasite viability. (Sujaan Das)

B1-O4 12:15 PM - 12:30 PM (15 mins)

Phosphoinositide metabolism links cGMP-dependent protein kinase G to essential Ca2+ signals at key decision points in the life cycle of malaria parasites (Mathieu Brochet)

Session C1 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment I - Chair: Alan Fairlamb, University of Dundee

C1-IS - Invited speaker Isabela Ribeiro, DNDi 11:00 AM - 11:30 AM (30 mins)

C1-O1 11:30 AM - 11:45 AM (15 mins)

Biochemical characterization and in vivo chemical validation of Trypanosoma brucei phosphodiesterases, as potential drug target (Srinivasa P S Rao)

C1-O2 11:45 AM - 12:00 PM (15 mins)

Targeting the nucleotide metabolism of Trypanosoma brucei I (Anders Hofer)

C1-O3 12:00 PM - 12:15 PM (15 mins)

Inhibitors of Leishmania major Inositol Phosphorylceramide Synthase – New Therapies for Leishmaniasis (Christopher Brown)

C1-O4 12:15 PM - 12:30 PM (15 mins)

A robust in vivo imaging model for late stage human African trypanosomiasis to evaluate antitrypanosomal drugs. (Hollie Burrell-Saward)

Session D1 (RCA) - Helminths- Diagnostics & Control - Chair: Joanne Webster, Imperial College London

D1-IS - Invited speaker Charlie King, 11:00 AM - 11:30 AM (30 mins)

Diagnosing 'Egg-negative' Schistosomiasis, a Modern Priority in the Move toward Elimination (Charles King)

D1-O1 11:30 AM - 11:45 AM (15 mins)

Schistosomiasis elimination in Zanzibar (Unguja and Pemba Islands): design and implementation of an integrated multidisciplinary research programme (Stefanie Knopp)

D1-O2 11:45 AM - 12:00 PM (15 mins)

The insulin receptor: an Achilles' heel for schistosome vaccine development (Donald McManus)

D1-O3 12:00 PM - 12:15 PM (15 mins)

Schistosomiasis in pre-school-aged children and their mothers in Chikhwawa district, southern Malawi with notes on the local freshwater snail fauna (Russell Stothard)

D1-O4 12:15 PM - 12:30 PM (15 mins)

A large-scale school based deworming programme in Bihar State, India – recipe for success. (Laura Appleby)

Session E1 (RCU) - Ecology - Bridging scales - Chair: Andy Fenton, University of Liverpool

E1-IS - Invited speaker Samuel Alizon, University of Montpellier 11:00 AM - 11:30 AM (30 mins)

Should we link within- and between-host levels in evolutionary epidemiology? (Samuel Alizon)

E1-O1 11:30 AM - 11:45 AM (15 mins)

Measuring health for parasitologists: Global Burden of Disease and beyond? (Thomas Fürst)

E1-O2 11:45 AM - 12:00 PM (15 mins)

Epidemiological feedbacks affect evolutionary emergence of pathogens (Matthew Hartfield)

E1-O3 12:00 PM - 12:15 PM (15 mins)

Do patterns of nestedness in parasite communities of wild wood mice predict order of infection in individual hosts? (Evelyn C. Rynkiewicz)

E1-O4 12:15 PM - 12:30 PM (15 mins)

Effective networking across metabolic, immune, and gut microbial systems may be crucial to fight Leishmania major infection (Sabrina Lamour)

Session F1 (RCJ) - Genomics of veterinary parasites - Chair: Matt Berriman (otherwise Fiona Tomley), Wellcome Trust Sanger Institute

F1-IS - Invited speaker John Gilleard, University of Calgary 11:00 AM - 11:30 AM (30 mins)

Genomic and genetic approaches to investigate the molecular basis of anthelmintic resistance: Haemonchus contortus as a model system (John Gilleard)

F1-O1 11:30 AM - 11:45 AM (15 mins)

Molecular epidemiology of Leishmania donovani in the Indian sub-continent: whole genome sequencing reveals bottlenecks, clonal outbreaks, migration and recombination (Jean-Claude Dujardin)

F1-O2 11:45 AM - 12:00 PM (15 mins)

VSG-Seq: A quantitative method for analyzing Trypanosoma brucei Variant Surface Glycoprotein expression in vivo (Monica Mugnier)

F1-O3 12:00 PM - 12:15 PM (15 mins)

Harnessing related species and samples data to create and optimise draft genome sequences for Leishmania species (Simone Coughlan)

F1-O4 12:15 PM - 12:30 PM (15 mins)

Application of Reverse Line Blotting to the study of tick-borne haemoparasites – A West African Experience (Vincenzo Lorusso)

Lunch

Lunch 12:30 PM - 2:00 PM (90 mins)

The Naked Scientists Radio Show recording from 1pm Robinson University

Radio 4 Science programme connecting the public with science 1:00 PM - 2:00 PM (60 mins)

Session A2 (WRL) - Kinetoplastida - Gene expression - Chair: Miguel Navarro, IPBLN - Spanish National Research Council

A2-IS - Invited speaker Isabel Roditi, University of Bern 2:00 PM - 2:30 PM (30 mins)

Gene expression in trypanosomes – the control freak's guide (Isabel Roditi)

A2-O1 2:30 PM - 2:45 PM (15 mins)

Chromatin readers regulate monoallelic expression and switching in Trypanosoma brucei. (Danae Schulz)

A2-O2 2:45 PM - 3:00 PM (15 mins)

The whole transcriptome analysis of Leishmania major virulence factors (Vyacheslav Yurchenko)

A2-O3 3:00 PM - 3:15 PM (15 mins)

Allelic exclusion by VEX1 controls antigenic variation in trypanosomes (Lucy Glover)

A2-O4 3:15 PM - 3:30 PM (15 mins)

A nucleolar protein of Trypanosoma brucei is involved in both surface protein and ribosomal RNA expression (Salome Aeschlimann)

Session B2 (WRR) - Apicomplexa - Mosquitoes and sexual stages - Chair: Jacob Baum, Walter & Eliza Hall Institute of Medical Research

B2-IS - Invited speaker Martin Donnelly , Liverpool School of Tropical Medicine 2:00 PM - 2:45 PM (45 mins)

B2-O1 2:45 PM - 3:05 PM (20 mins)

Role of the apiAP2 proteins in the life cycle of malaria parsite. (Katarzyna Modrzynska)

B2-O2 3:05 PM - 3:25 PM (20 mins)

High throughput reverse genetics screening in Plasmodium berghei using Signature tagged mutagenesis unravels genetic interactions (Ana Rita Gomes)

Session C2 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment II - Chair: Jeremy Sternberg, University of Aberdeen

C2-IS - Invited speaker Michael Miles, London School of Hygiene and Tropical Medicine 2:00 PM - 2:30 PM (30 mins)

Chagas disease: progress and challenges" (Michael Miles)

C2-O1 2:30 PM - 2:45 PM (15 mins)

Global Distribution Maps of the Leishmaniases (David Pigott)

C2-O2 2:45 PM - 3:00 PM (15 mins)

Trypanosomiasis in domestic livestock in the Luangwa valley in Zambia. (Kathrin Schaten)

C2-O3 3:00 PM - 3:15 PM (15 mins)

Mapping the incidence and detection probability of human African trypanosomiasis (Nick Golding)

C2-O4 3:15 PM - 3:30 PM (15 mins)

Human migration drives the dispersal of epizootic Chagas disease: the case of highland Bolivia (Louisa Messenger)

Session D2 (RCA) - Helminths - Molecular epidemiology and transmission - Chair: David Rollinson, The Natural History Museum

D2-IS - Invited speaker Sam Loker, University of New Mexico 2:00 PM - 2:30 PM (30 mins)

Achieving Transmission Control of Schistosomiasis: Is There a Role for Biological Enemies of Snails or Schistosomes? (Eric Loker)

D2-O1 2:30 PM - 2:45 PM (15 mins)

Population genetic studies of Schistosoma haematobium using novel multiplex microsatellites: inter and intra species molecular epidemiology (Bonnie Webster)

D2-O2 2:45 PM - 3:00 PM (15 mins)

Distribution of Tetraspanin-23 alleles in hybrid schistosomes. (Tine Huyse)

D2-O3 3:00 PM - 3:15 PM (15 mins)

Look to the snails: indicators of schistosomiasis transmission within control programmes (Fiona Allan)

D2-O4 3:15 PM - 3:30 PM (15 mins)

Pattern of soil-transmitted helminth re-infections among Orang Asli schoolchildren in Malaysia (Hesham Al-Mekhlafi)

Session E2 (RCU) - Ecology - Local adaptation and coevolution - Chair: Olivier Restif, University of Cambridge

E2-IS - Invited speaker Oliver Kaltz, Université Montpellier 2 2:00 PM - 2:30 PM (30 mins)

Environmental stochasticity affects epidemics and host-parasite coevolution (Oliver Kaltz)

E2-O1 2:30 PM - 2:45 PM (15 mins)

Host colour change and scent affect survival of the nematode Heterorhabditis bacteriophora (Rebecca Jones)

E2-O2 2:45 PM - 3:00 PM (15 mins)

Assessment and Management of emerging nematode pests of Northern Ireland grassland and cereals (Thomas Fleming)

E2-O3 3:00 PM - 3:15 PM (15 mins)

Bayesian modelling of factors potentially influencing the spatial distribution of Echinococcus multilocularis in foxes (Franz J. Conraths)

E2-O4 3:15 PM - 3:30 PM (15 mins)

Predatory capacity of different copepods against Aedes aegypti larvae from Lahore (Nusrat Jahan)

Session F2 (RCJ) - One Health - Chair: Diana Williams, University of Liverpool

F2-IS - Invited speaker Paul Torgerson, University of Zurich 2:00 PM - 2:30 PM (30 mins)

One health: parasites and priorities. (Paul Torgerson)

F2-O1 2:30 PM - 2:45 PM (15 mins)

Migration and the risk of animal trypanosomiasis on the Jos Plateau, Nigeria (Oluwashola Olaniyan)

F2-O2 2:45 PM - 3:00 PM (15 mins)

Arsenic, antimony and Leishmania – has arsenic contamination of drinking water in India led to treatment resistant kala-azar? (Meghan Perry)

F2-O3 3:00 PM - 3:15 PM (15 mins)

WIPO Re:Search – A Catalyst for Success: Channeling the Expertise of Industry, Academic, and Nonprofit Organizations toward Neglected Disease Research (Katy Graef)

F2-O4 3:15 PM - 3:30 PM (15 mins)

Afternoon Refreshments

Coffee and Tea Break 3:30 PM - 4:00 PM (30 mins)

Session A3 (WRL) - Kinetoplastida - RNA biology - Chair: C Clayton, University of Heidelberg

A3-IS - Invited speaker Elisabetta Ullu, Yale 4:00 PM - 4:30 PM (30 mins)

Moving forward: from procyclics to metacyclics in Trypanosoma brucei (Elisabetta Ullu)

A3-O1 4:30 PM - 4:45 PM (15 mins)
Purification of specific mRNPs via the nascent polypeptide (Diana Patricia Inchaustegui Gil)

A3-O2 4:45 PM - 5:00 PM (15 mins)
Comparative ribosome-profiling reveals extensive translational complexity in different Trypanosoma brucei life-cycle stages (Juan José Vasquez)

A3-O3 5:00 PM - 5:15 PM (15 mins)
Post-transcriptional control of mRNAs by ZC3H11 and MKT1 (Dorothea Droll)

A3-O4 5:15 PM - 5:30 PM (15 mins)
Transcriptome-wide analysis of mRNA decay in trypanosomes reveals complex degradation kinetics and novel control mechanisms (Christine Clayton)

Session B3 (WRR) - Apicomplexa - Biochemistry - Chair: Oliver Billker, Wellcome Trust Sanger Institute

B3-IS - Invited speaker Moritz Treeck, MRC National Institute for Medical Research

4:00 PM - 4:30 PM (30 mins)
Using quantitative phosphoproteome and proteome analysis to identify signaling pathways controlled by parasitic kinases (Moritz Treeck)

B3-O1 4:30 PM - 4:45 PM (15 mins)
Comparative metabolomics of erythroid lineage: implications for malaria control (Anubhav Srivastava)

B3-O2 4:45 PM - 5:00 PM (15 mins)
Polycistronic and antisense transcription of the Plasmodium apicoplast genome (Ellen Nisbet)

B3-O3 5:00 PM - 5:15 PM (15 mins)
Development of a directional, amplification-free RNA-seq protocol optimised for AT-rich Plasmodium parasites (Lia Chappell)

B3-O4 5:15 PM - 5:30 PM (15 mins)
The antimalarial action of FK506, rapamycin and non-immunosuppressive congeners: evidence for a direct effect on FK506-binding protein (Angus Bell)

Session C3 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment III - Chair: Sanjeev Krishna, St George's, University of London

C3-IS - Invited speaker Paul Kaye, University of York 4:00 PM - 4:30 PM (30 mins)

Pathways to Leishmania persistence: in vivo veritas? (Paul Kaye)

C3-O1 4:30 PM - 4:45 PM (15 mins)
Bioluminescence imaging of chronic Trypanosoma cruzi infections reveals tissue-specific parasite dynamics and heart disease in the absence of locally persistent infection (Amanda Fortes Francisco)

C3-O2 4:45 PM - 5:00 PM (15 mins)
Aquaporin 2 is the main determinant for pentamidine and melaminophenyl arsenical resistance in Trypanosoma brucei spp. (Harry De Koning)

C3-O3 5:00 PM - 5:15 PM (15 mins)
Host immunity is regulated by alternatively activated macrophages in Indian Post Kala-Azar Dermal Leishmaniasis (Mitali Chatterjee)

C3-O4 5:15 PM - 5:30 PM (15 mins)
Leishmania mexicana modulates dendritic cell migration towards draining lymph nodes (Jenny Crowe)

Session D3 (RCA) - Helminths - Human immunology and morbidity - Chair: Shona Wilson, University of Cambridge

D3-IS1 - Invited speaker Birgitte Vennervald, University of Copenhagan 4:00 PM - 4:30 PM (30 mins)

D3-O1 4:30 PM - 4:45 PM (15 mins)
Identifying and quantifying morbidity markers associated with Schistosoma haematobium infection in children. (Welcome Mkululi Wami)

D3-O2 4:45 PM - 5:00 PM (15 mins)
Anti-glycan antibody responses upon infection or vaccination with Schistosoma mansoni (Angela van Diepen)

D3-O3 **5:00 PM - 5:15 PM (15 mins)**

IgE responses to abundant antigens in the parasite Schistosoma mansoni: A link between allergy and the evolved immune response to metazoan parasites? (Edward Farnell)

D3-O4 **5:15 PM - 5:30 PM (15 mins)**

High throughput-compatible identification of novel helminth allergens using a humanised basophil reporter cell line (Franco Falcone)

Session E3 (RCU) - Ecology - Disease transmission - Chair: Sarah Perkins, Cardiff University

E3-IS - Invited speaker Elizabeth Murchison, University of Cambridge **4:00 PM - 4:30 PM (30 mins)**

Genome analysis of clonally transmissible cancers in dogs and Tasmanian devils (Elizabeth Murchison)

E3-O1 **4:30 PM - 4:45 PM (15 mins)**

A not so slow boat to China: an update on the story of Biomphalaria, invasive carriers of neotropical schistosomiasis, in China (Stephen Attwood)

E3-O2 **4:45 PM - 5:00 PM (15 mins)**

Epidemiology of Human Leptospirosis in Malaysia (2004 -2012) (Mohd Zain Siti Nursheena)

E3-O3 **5:00 PM - 5:15 PM (15 mins)**

The consequences of coinfections for parasite transmissionin in the mosquito Aedes aegypti (Alison Duncan)

E3-O4 **5:15 PM - 5:30 PM (15 mins)**

Transmission dynamics of pathogenic bacteria by free-living nematodes (Anaid Diaz)

Session F3 (RCJ) - Parasite epidemiology - Chair: Paul Torgerson, University of Zurich

F3-IS - Invited speaker Eric Morgan, RVC **4:00 PM - 4:30 PM (30 mins)**

Climate change and the epidemiology of nematode parasites: issues of scale (Eric Morgan)

F3-O1 **4:30 PM - 4:45 PM (15 mins)**

Immuno-epidemiological models predict novel markers for parasite resistance (Joaquín Prada Jiménez de Cisneros)

F3-O2 **4:45 PM - 5:00 PM (15 mins)**

Transmission dynamics and control of Fasciola hepatica within sheep in the UK: the impact of population structure (Nicola Beesley)

F3-O3 **5:00 PM - 5:15 PM (15 mins)**

Quantifying the effects of individual animal characteristics and climatological factors on faecal worm egg count shedding in donkeys (Christopher Corbett)

F3-O4 **5:15 PM - 5:30 PM (15 mins)**

Tick borne pathogens in Nigerian livestock; the influence of acaricide treatment on tick burden (Joe Farrimond)

The African experience (WRA): Prof David Dunne

The African experience (WRA): Prof David Dunne **5:30 PM - 6:00 PM (30 mins)**

The Africa Experience (David Dunne)

Poster sessiom (WRA)

Posters and Drinks **6:00 PM - 8:00 PM (120 mins)**

Young parasitologist's party (The Avery, Regent Street)

More Drinks and Quiz. **8:00 PM - 11:59 PM (239 mins)**

Registration - Tuesday 8th April

Registration: West Road **8:30 AM - 5:30 PM (540 mins)**

Plenary session 4 (WRA):

David Roos - University of Pennsylvania **9:00 AM - 9:30 AM (30 mins)**

CA Wright medal lecture (WRA): Michael Barrett, University of Glasgow & Alex Rowe, University of Edinburgh

CA Wright medal lecture - Michael Barrett, University of Glasgow 9:30 AM - 10:00 AM (30 mins)

Modes of action and mechanisms of resistance to anti-protozoal drugs (Michael Barrett)

CA Wright medal lecture - Alex Rowe, University of Edinburgh 10:00 AM - 10:30 AM (30 mins)

Investigating a 100 year-old mystery: receptor-ligand interactions in severe malaria (Alex Rowe)

Morning Refreshments

Coffee and Tea Break 10:30 AM - 11:00 AM (30 mins)

Session A4 (WRA) - Kinetoplastida - Growth, form and metabolism I - Chair: Eva Gluenz, University of Oxford

A4-IS - Invited speaker Markus Engstler, Universität Würzburg 11:00 AM - 11:30 AM (30 mins)

Expression site attenuation and development in Trypanosoma brucei (Markus Engstler)

A4-O1 11:30 AM - 11:45 AM (15 mins)

The trypanosome bilobe: form, fabric, and function (Brooke Morriswood)

A4-O2 11:45 AM - 12:00 PM (15 mins)

The transmembrane protein homologue GPR89 promotes the development of stumpy forms in Trypanosoma brucei (Federico Rojas)

A4-O3 12:00 PM - 12:15 PM (15 mins)

VSG identity and structural integrity determine growth rate of bloodstream form Trypanosoma brucei (Angela Schwede)

A4-O4 12:15 PM - 12:30 PM (15 mins)

Uncoupling flagellum formation and maintenance (Cecile Fort)

Session B4 (WRR) - Apicomplexa - Translation - Chair: Julian Rayner, Wellcome Trust Sanger Institute

B4-IS - Invited speaker Simon Draper , University of Oxford 11:00 AM - 11:30 AM (30 mins)

Towards the Development of a Broadly-Neutralising Vaccine against Blood-Stage Plasmodium falciparum. (Simon Draper)

B4-O1 11:30 AM - 11:45 AM (15 mins)

Development of Transgenic Rodent Malaria Parasites for Assessment of Novel Liver-Stage Malaria Vaccines (Ahmed M. Salman)

B4-O2 11:45 AM - 12:00 PM (15 mins)

Efficacy of dihydroartemisinin-piperaquine in asymptomatic malaria parasite carriers: an evaluation of molecular markers of drug resistance (Mary C. Oguike)

B4-O3 12:00 PM - 12:15 PM (15 mins)

Emetine dihydrochloride hydrate: a potential candidate for repositioning in malaria (Holly Matthews)

B4-O4 12:15 PM - 12:30 PM (15 mins)

Session C4 (WRL) - Evolution I - Chair: Mark Field,

C4-IS - Invited speaker Julius Lukas , University of Southern Bohemia 11:00 AM - 11:30 AM (30 mins)

Diversity and phylogeny of insect trypanosomatids (Julius Lukes)

C4-O1 11:30 AM - 11:45 AM (15 mins)

Evolution of chloroplast RNA processing at the boundary between photosynthetic and parasitic apicomplexans (Richard Dorrell)

C4-O2 11:45 AM - 12:00 PM (15 mins)

Applied evolution: an experimental approach investigating how drug dosage affects the rate of resistance evolution. (Alan Reynolds)

C4-O3 12:00 PM - 12:15 PM (15 mins)

Expansion and sculpting of the membrane trafficking system in the neuropathogenic amoeba Naegleria fowleri (Emily Herman)

C4-O4 12:15 PM - 12:30 PM (15 mins)
Streamlined endocytosis in Trypanosoma brucei (Paul Manna)

Session D4 (RCA) - Helminths - Genomic, Proteomic & Glycomic Biology - Chair: Cornelis Hokke, University of Lieden

D4-IS - Invited speaker Peter Geldhof, University of Ghent 11:00 AM - 11:30 AM (30 mins)
How do 'omics' technologies help us to control nematode infections? (Peter Geldhof)

D4-O1 11:30 AM - 11:45 AM (15 mins)
Fasciola hepatica: in vitro maintenance and evaluation of unique tegumental proteins as novel control targets (Paul McCusker)

D4-O2 11:45 AM - 12:00 PM (15 mins)
Schistosoma mansoni methyl-CpG binding domain protein (SmMBD2/3): a functional component of the schistosome epigenetic machinery (Sabrina Munshi)

D4-O3 12:00 PM - 12:15 PM (15 mins)
Characterization of excretory/secretory products and small metabolites of the pig whipworm Trichuris suis: insights on immunomodulation by intestinal parasites (Louis-Philippe Leroux)

D4-O4 12:15 PM - 12:30 PM (15 mins)
The whipworm genome and transcriptome (Bernardo Foth)

Session E4 (RCU) - Public understanding, outreach and communication - Presenting Science to the Public - On Their Terms -
Chair: Tansy Hammarton, University of Glasgow

E4-IS - Invited speaker Michael Barrett, University of Glasgow 11:00 AM - 11:30 AM (30 mins)
Presenting science to the public - on their terms (Mike Barrett)

E4-O1 11:30 AM - 11:45 AM (15 mins)
Crafty Critters: crafting a parasite infestation ((1) Rebecca (2) Catarina A (1) Devlin (2) Marques)

E4-O2 11:45 AM - 12:00 PM (15 mins)
Communicating science to the public across the generations (Linda KOHL)

E4-O3 12:00 PM - 12:30 PM (30 mins)
Tapeworm Diaries (Philip Craig)

Session F4 (RCJ) - Antihelmintic overuse - evolution under pressure - Chair: Jacqui Matthews, Moredun

F4-IS - Invited speaker Andrew Read, Penn State 11:00 AM - 11:15 AM (15 mins)

F4-O1 11:30 AM - 11:45 AM (15 mins)
The emergence of anthelmintic resistance in parasitic nematodes of livestock is characterised by multiple hard and soft selective sweeps of independently derived mutations (Libby Redman)

F4-O2 11:45 AM - 12:00 PM (15 mins)
Help: the wormers don't work! (Jacqueline Matthews)

F4-O3 12:00 PM - 11:15 AM (-45 mins)
Metabolic enzymes of the liver fluke, Fasciola hepatica: biochemical characterisation and identification of inhibitors (David Timson)

F4-O4 12:15 PM - 12:30 PM (15 mins)
Pharmacokinetic/Pharmacodynamic modelling of anti-Wolbachia agents. (David Waterhouse)

Lunch

Lunch 12:30 PM - 2:00 PM (90 mins)

Social media and crowdsourcing - Dane Comerford, Public Engagement Manager, at University of Cambridge (RobU) Social media and crowdsourcing 1:00 PM - 2:00 PM (60 mins)

Session A5 (WRA) - Kinetoplastida - Growth, form and metabolism II - Chair: Mike Barrett, University of Glasgow

A5-IS - Invited speaker Frederic Bringaud, Université Bordeaux Segalen 2:00 PM - 2:30

PM (30 mins)

Comparison of the central metabolism of the insect and bloodstream trypanosomes (Frédéric Bringaud)

A5-O1 2:30 PM - 2:45 PM (15 mins)

Iron uptake in Trypanosoma Brucei (Martin Taylor)

A5-O2 2:45 PM - 3:00 PM (15 mins)

Variations in Swimming Patterns and Behaviour of African Trypanosomes in Mammalian Hosts Depicts Adaptation to Survive in Diverse Environments (Joel Bargul)

A5-O3 3:00 PM - 3:15 PM (15 mins)

Developing a flagellar transition zone proteome in Trypanosoma brucei (Samuel Dean)

A5-O4 3:15 PM - 3:30 PM (15 mins)

The surface landscape of African trypanosomes (Catarina Gadelha)

Session B5 (WRR) - Apicomplexa - Pathogenesis - Chair: Catherine Merrick, Keele University

B5-IS - Invited speaker Matthew Higgins , University of Oxford 2:00 PM - 2:30 PM (30 mins)

Conserved structural features required for interaction with endothelial protein C receptor in severe malaria (Matthew Higgins)

B5-O1 2:30 PM - 2:45 PM (15 mins)

Alteration of the Blood-Brain Barrier (BBB) endothelial cells, secondary to Plasmodium falciparum infected red blood cells (PRBC) sequestration in Cerebral Malaria: an in-vitro study (Mohd Hamzah Mohd Nasir)

B5-O2 2:45 PM - 3:00 PM (15 mins)

Generation of antigenic diversity in Plasmodium falciparum by structured rearrangement of var genes (William Hamilton)

B5-O3 3:00 PM - 3:15 PM (15 mins)

A systems biology approach to characterizing host-parasite interactions during Toxoplasma cell invasion (Dong Xia)

B5-O4 3:15 PM - 3:30 PM (15 mins)

Strain-transcending antibodies against group A PfEMP1 variants implicated in severe childhood Plasmodium falciparum malaria (Ashfaq Ghumra)

Session C5 (WRL) - Evolution II - Chair: Jamie Stevens, University of Exeter

C5-IS - Invited speaker Joel Dacks , University of Alberta 2:00 PM - 2:30 PM (30 mins)

From endolysosomes to invasion organelles: evolutionary remodeling of membrane-trafficking across the Apicomplexa. (Joel Dacks)

C5-O1 2:30 PM - 2:45 PM (15 mins)

Evolution of the nuclear pore complex (Mark Field)

C5-O2 2:45 PM - 3:00 PM (15 mins)

Architecture and Evolution of the Trypanosome Nuclear Pore Complex (SAMSON OBADO)

C5-O3 3:00 PM - 3:15 PM (15 mins)

Identification of a novel adaptin-related coat complex (Alexander Schlacht)

C5-O4 3:15 PM - 3:30 PM (15 mins)

NUP-2, a second component of the trypanosome nucleoskeleton (Luke Maishman)

Session D5 (RCA) - Helminths - Co-infections and transmission dynamics - Chair: Andrea Graham, Princeton University

D5-IS - Invited speaker Andrea Graham , Princeton University 2:00 PM - 1:30 PM (-30 mins)

Dynamics of helminth-microparasite co-infections, from pairwise interactions in the lab to realistic complexity in the field (Andrea Graham)

D5-O1 2:30 PM - 2:45 PM (15 mins)

Effects of parasite interactions on the survival of a wild rodent (Godefroy Devevey)

D5-O2 2:45 PM - 3:00 PM (15 mins)

Female host sex-biased parasitism with Mastophorus muris in wild bank voles (Myodes glareolus). (Grzybek Maciej)

D5-O3 3:00 PM - 3:15 PM (15 mins)

Costs of resistance and costs of infections on the fitness of the mosquito Aedes aegypti infected

with the filarial nematode Brugia malayi (Cristina Ariani)

D5-O4 3:15 PM - 3:30 PM (15 mins)

Onchocerciasis transmission in Ghana: the effect of simuliid cytospecies and host blood-meal choice (Poppy Lamberton)

Session E5 (RCU) - Ecology - Aquatic Parasitology & Sustainable Fisheries, in memory of Professor Angela Davie - Chair: Rachel Norman, University of Stirling

Brief history of Angela Davie 1:59 PM - 2:00 PM (1 mins)

This session is dedicated to the memory of Professor Angela Davies (Russell) (Angela Davie)

E5-IS - Invited speaker Nick Taylor, Cefas 2:00 PM - 2:30 PM (30 mins)

Pathogen control in aquatic systems: A national challenge (Nick Taylor)

E5-O1 2:30 PM - 2:45 PM (15 mins)

Parasitic or not? Symbiotic branchiobdellids (Annelida: Clitellata) on invasive signal crayfish (Pacifastacus leniusculus) (Jo James)

E5-O2 2:45 PM - 3:00 PM (15 mins)

Why does maternal food matter? Maternal effects on body size modify disease resistance in Daphnia magna offspring. (Jennie Garbutt)

E5-O3 3:00 PM - 3:15 PM (15 mins)

Changes in parasite within-host dynamics and estimates of genetic variation over time (Melanie Clerc)

E5-O4 3:15 PM - 3:30 PM (15 mins)

Parasites of Trinidadian guppies, Poecilia reticulata: evidence for sex- and age-specific traitmediated indirect effects of predators (Jessica Stephenson)

Session F5 (RCJ) - Careers - Careers Workshop - Chair: Emily Dawson & Luke Roberts

F5-IS - Invited speaker Mike Turner, Wellcome Trust 2:00 PM - 2:15 PM (15 mins)

Wellcome Trust fellowship schemes (Mike Turner)

F5-IS - Invited speaker Emily Adams, Liverpool School of Tropical Medicine 2:15 PM - 2:30 PM (15 mins)

Life as a junior academic (Emily Adams)

F5-IS - Invited speaker Mike Leahy, TV Presenter Oxford University 2:30 PM - 2:45 PM (15 mins)

Career paths and the bizarre challenges along the way (Mike Leahy)

F5-IS - Invited speaker Lesley Drake, Imperial College London 2:45 PM - 3:00 PM (15 mins)

How and why you should pursue various career options (Leslie Drake)

F5-Wo - Workshop Q & A 3:00 PM - 3:30 PM (30 mins)

Afternoon Refreshments

Coffee and Tea Break 3:30 PM - 4:00 PM (30 mins)

Session A6 (WRA) - Kinetoplastida - Interactions with vertebrate and arthropod hosts - Chair: Mark Carrington, University of Cambridge

A6-IS - Invited speaker Steve Beverley, Washington University 4:00 PM - 4:30 PM (30 mins)

Leishmania proteophosphoglycans (PPGs): genetic analysis of a parasite mucin-like glycoprotein implicated in several key steps in the infectious cycle (Stephen Beverley)

A6-O1 4:30 PM - 4:45 PM (15 mins)

Unzipping the barriers: how trypanosomes breach the tsetse peritrophic matrix (Clair Rose)

A6-O2 4:45 PM - 5:00 PM (15 mins)

Both host and parasite genetic factors determine long-term tissue-specific infection dynamics in experimental chronic Chagas disease (Michael Lewis)

A6-O3 5:00 PM - 5:15 PM (15 mins)

In search of anti-disease vaccine candidates for African trypanosomiasis: new insights into the protein composition of trypanosome-infected tsetse saliva (Alvaro Acosta-Serrano)

A6-O4 5:15 PM - 5:30 PM (15 mins)

Novel method for quantifying Leishmania metacyclics promastigotes delivered by sand fly bite.

(Matthew Rogers)

Session B6 (WRR) - Apicomplexa - Other Apicomplexa - Chair: Janet Cox-Singh, University of St Andrews

B6-IS - Invited speaker Colin Sutherland, London School of Hygiene and Tropical Medicine 4:00 PM - 4:30 PM (30 mins)

Adaptive biology of the persistent human parasites Plasmodium malariae, P. ovale curtisi and P. ovale wallikeri (Colin Sutherland)

B6-O1 4:30 PM - 4:45 PM (15 mins)

Structure, Evolution and Function of the Genome of the Emerging Human Pathogen Babesia microti. (Emmanuel Cornillot)

B6-O2 4:45 PM - 5:00 PM (15 mins)

Whole Genome Amplification from clinical samples: Advances in Cryptosporidium genome sequencing from limited numbers of oocysts (Jenna Alexander)

B6-O3 5:00 PM - 5:15 PM (15 mins)

Plasmodium knowlesi - another malaria parasite. (Janet Cox-Singh)

B6-O4 5:15 PM - 5:30 PM (15 mins)

The European Malaria Reagent Repository – the Rodent Malaria Collection. (Joanne Thompson)

Session C6 (WRL) - Vectors and Parasites in the Middle East - Vectors and Parasites in the Middle East I - Chair: Ashraf Mohamed Ahmed Ali, King Saud University

C6-IS - Invited speaker Saeed Alharthi, Umm Al-Qura University, Saudi Arabia 4:00 PM - 4:30 PM (30 mins)

Malaria in Saudi Arabia, the current situation. (Saeed Al-Harthi)

C6-O1 4:30 PM - 4:45 PM (15 mins)

Hydatidosis and Echinococcosis in Gaza strip (Adnan Al-Hindi)

C6-O2 4:45 PM - 5:00 PM (15 mins)

Morphological and Phylogenetic analysis of Serrasentis sagittifer (Acanthocephala: Rhadinorhynchidae) isolated from the Gilthead Sea bream Sparus aurata (Sparidae) , Red Sea, Egypt (Rewaida Abdel-Gaber)

C6-O3 5:00 PM - 5:15 PM (15 mins)

Kudoa parasites infecting oocytes : characterization of new species arguing in favor of the induction of a xenoma-like structure (Lamjed Mansour)

C6-O4 5:15 PM - 5:30 PM (15 mins)

~~Molecular characterization of Stictodora tridactyla (Digenea: Heterophyidae) using ITS1 and mtCO1 sequences in Kuwait (Wafa Al-Kandari)~~

Session D6 (RCA) - Helminths - Experimental Immunology - Mini Symposium - Chair: Richard Grencis/Andrew MacDonald, Manchester University

D6-IS1 - Invited speaker Mark Wilson , NIMR, London 4:00 PM - 4:20 PM (20 mins)

D6-IS2 - Invited speaker Rick Maizels , Edinburgh 4:20 PM - 4:40 PM (20 mins)

D6-IS3 - Invited speaker Richard Grencis, Manchester University 4:40 PM - 5:00 PM (20 mins)

D6-IS4 - Invited speaker Andrew MacDonald, Manchester University 5:00 PM - 5:20 PM (20 mins)

D6-Q1 - Discussion 5:20 PM - 5:30 PM (10 mins)

Session E6 (RCU) - Ecology - Global Weirding, invasive species and parasitism - Chair: Jo Cable, Cardiff University

E6-IS - Invited speaker Kate Jones, University College London 4:00 PM - 4:30 PM (30 mins)

Predicting the global emergence and spread of zoonotic infectious diseases (Kate Jones)

E6-O1 4:30 PM - 4:45 PM (15 mins)

Predicting the effects of climate change on Schistosoma mansoni transmission in East Africa: A mathematical modelling study (Nicky McCreesh)

E6-O2 4:45 PM - 5:00 PM (15 mins)

Distribution and abundance of Ixodid ticks as vectors of disease in Northern Ireland. (Jonathan Lappin)

E6-O3 5:00 PM - 5:15 PM (15 mins)

Seasonal dynamics and long term trends in a host-parasite community (Caroline Millins)

E6-O4 5:15 PM - 5:30 PM (15 mins)

The effect of climate perturbations on parasite life-history variables (Emma Gillingham)

Session F6 (RCJ) - Emerging Vaccines - Chair: Al Nisbet, Moredun Research Institute

F6-IS1 - Invited speaker Tom McNeilly, Moredun Research Institute 4:00 PM - 4:30 PM (30 mins)

Development of a subunit nematode vaccine: antigen discovery, antigen generation and adjuvant development (Tom McNeilly)

F6-IS2 - Invited speaker John Dalton, Queens University Belfast 4:30 PM - 5:00 PM (30 mins)

Fasciola hepatica cathepsin L vaccines: we're not there…yet!

F6-O1 5:00 PM - 5:15 PM (15 mins)

Population, genetic and antigenic diversity of Eimeria: prospects for novel vaccines (Damer Blake)

F6-O2 5:15 PM - 5:30 PM (15 mins)

Characterising immune responses in UK dairy cattle naturally exposed to Fasciola hepatica (John Graham-Brown)

Conference Dinner

Conference Gala Dinner 7:00 PM - 12:00 AM (-1140 mins)

Registration - Wednesday 9th April

Registration: West Road 8:30 AM - 12:00 PM (210 mins)

BSP debate (WRA): Vector control is too environmentally damaging for its beneficial public health impact: yes or no? with Prof Russ Stothard, followed by AGM

BSP debate, EGM and AGM 9:00 AM - 10:30 AM (90 mins)

Morning Refreshments

Coffee and Tea Break 10:30 AM - 11:00 AM (30 mins)

Session A7 (WRA) - Kinetoplastida - Respond to Abstracts - Chair: Mark Field, University of Dundee

A7-O1 11:00 AM - 11:15 AM (15 mins)

Novel components of the mitochondrial segregation machinery and their hierarchy uncovered in Trypanosoma brucei (Torsten Ochsenreiter)

A7-O2 11:15 AM - 11:30 AM (15 mins)

Discovering single nucleotide polymorphisms and structural variations in homogeneous and heterogeneous populations of trypanosomatids (Hideo Imamura)

A7-O3 11:30 AM - 11:45 AM (15 mins)

Using NextGen data to model kDNA segregation and predict guide RNA genes in Trypanosoma brucei (Sinclair Cooper)

A7-O4 11:45 AM - 12:00 PM (15 mins)

The effect of Leishmania major infection on atherogenesis and cytokines pattern in resistant and susceptible mice (Marc Karam)

A7-O6 12:15 PM - 12:30 PM (15 mins)

The dynamics of mitochondrial RNA binding complex in Trypanosoma brucei and its petite mutant under optimized immobilization conditions (Hassan Hashimi)

A7-O5 1:00 PM - 1:15 PM (15 mins)

What do kinetoplastids need a kinetoplast for? Life cycle progression of Trypanosoma brucei in the presence and absence of mitochondrial DNA (Caroline Dewar)

Session B7 (WRR) - Apicomplexa - Immunology - Chair: Julius Hafalla, London School of Hygiene and Tropical Medicine

B7-IS - Invited speaker Jean Langhorne, MRC National Institute for Medical Research
11:00 AM - 11:30 AM (30 mins)

Immune responses and virulence in Plasmodium chabaudi infection (Jean Langhorne)

B7-O1 11:30 AM - 11:45 AM (15 mins)

Localising Selection from Resequencing Data: Linking Genes to Phenotypes in Malaria Parasites (Chris Illingworth)

B7-O2 11:45 AM - 12:00 PM (15 mins)

A comparative transcriptomic and proteomic investigation of host cell responses during Toxoplasma gondii and Neospora caninum invasion of human astrocytes. (Sarah Altwaim)

B7-O3 12:00 PM - 12:15 PM (15 mins)

Parasitology and inflammation in kidneys and lungs in a murine model of co-infection Plasmodium/filarial nematode (Gregory KARADJIAN)

B7-O4 12:15 PM - 12:30 PM (15 mins)

No Evidence that Knops Blood Group Polymorphisms Affect Complement Receptor 1 Clustering on Erythrocytes (Olivia Swann)

Session C7 (WRL) - Vectors and Parasites in the Middle East - Vectors and Parasites in the Middle East II - Chair: Mahmoud N. Abo- Shehada, London School of Hygiene and Tropical Medicine

C7-IS - Invited speaker Fathy A. Abdel-Ghaffar, Cairo University 11:00 AM - 11:30 AM (30 mins)

Sarcosporidia and Sarcosporidiosis (Apicomplexa: Coccidia) infecting reptiles in Egypt and Saudia Arabia (Fathy Abdel-Ghaffar)

C7-O1 11:30 AM - 11:45 AM (15 mins)

Parasitology research in the Middle East and North Africa 1950-2013 (Mahmoud Abo-Shehada)

C7-O2 11:45 AM - 12:00 PM (15 mins)

Bacillus thuringiensis Induces Cellular Stress in the Mosquito Vector, Culex pipiens, Prior to Death (Ashraf Ahmed)

C7-O3 12:00 PM - 12:15 PM (15 mins)

Insecticidal Activity of Newly Isolated Actinomycete Strains from the Desert Habitats of Saudi Arabia Against Culex pipiens (Wael Hozzein)

C7-O4 12:15 PM - 12:30 PM (15 mins)

Dynamic trends in intestinal parasitic infections among recently arrived immigrant workers, settled immigrants and long-term residents in Qatar. (Jerzy Behnke)

Session D7 (RCA) - Helminths - Helminths - Chemotherapy & Drug Targets - Chair: Russell Stodhard, Liverpool School of Tropical Medicine

D7-IS - Invited speaker Jutta Reinhard-Rupp, Merck Serono 11:00 AM - 11:30 AM (30 mins)

Schistosomiasis and Praziquantel – past and future of a gold standard chemotherapy (Jutta Reinhard-Rupp)

D7-O1 11:30 AM - 11:45 AM (15 mins)

Oxantel pamoate against Trichuris trichiura infections (Benjamin Speich)

D7-O2 11:45 AM - 12:00 PM (15 mins)

Molecular diagnosis of anthelmintic resistance in parasitic nematodes (Roger Prichard)

D7-O3 12:00 PM - 12:15 PM (15 mins)

A•WOL macrofilaricidal drug discovery and development - optimisation of anti-Wolbachia efficacy (Louise Ford)

D7-O4 12:15 PM - 12:30 PM (15 mins)

Bridging wet and dry labs: a proof of concept for rational drug design against tropical diseases (Adriana Erica Miele)

Day 4 Session E7 (RCU) - Ecology - Genetics, Evolution and Ecology of Host-Parasite Interaction - Chair: Ben Longdon,

E7-IS - Invited speaker Matthew Fisher, Imperial College London 11:00 AM - 11:30 AM (30 mins)

Global bio-insecurity breaches phylogeographic barriers leading to panzootic amphibian/chytrid parasitisms (Matthew Fisher)

E7-O1 11:30 AM - 11:45 AM (15 mins)

Why do flies vary in their susceptibility to infection? (Frank Jiggins)

E7-O2 11:45 AM - 12:00 PM (15 mins)

Covert specialism of apparently shared Bartonella parasites within woodland rodent communities. (Susan Withenshaw)

E7-O3 12:00 PM - 12:15 PM (15 mins)

E7-O4 12:15 PM - 12:30 PM (15 mins)

An experimentally evolved trypanosome and its implication for infection success and virulence in the bumblebee Bombus terrestris (Monika Marxer)

Session F7 (RCJ) - Food Security - Chair: Damer Blake, Royal Veterinary College

F7-IS - Invited speaker Katharina Staerk, Royal Veterinary College 11:00 AM - 11:30 AM (30 mins)

Parasites and Food Security: A complex relationship (Katharina Stärk)

F7-O1 11:30 AM - 11:45 AM (15 mins)

Farmer control of gastrointestinal parasites: why they do what they do? (Jacques Cabaret)

F7-O2 11:45 AM - 12:00 PM (15 mins)

Aquatic Food security- the role of parasites in seafood production. (Rachel Norman)

F7-O3 12:00 PM - 12:15 PM (15 mins)

Forecasting Nematodirus battus disease incidence by modelling known hatching dynamics (Owen Gethings)

F7-O4 12:15 PM - 12:30 PM (15 mins)

Improved use of abattoir information to aid the management of liver fluke in cattle and sheep (Stella Mazeri)

Lunch meeting close

Lunch 12:30 PM - 2:00 PM (90 mins)

Public Understanding of Science Lecture

West Road Auditorium, 5:30pm Monday 7th April

The Africa Experience

David Dunne, University of Cambridge

In the late 17th century, self-interest in the scramble for Africa crucially motivated UK infectious tropical disease research in sub-Saharan Africa. In the current era of more philanthropic support for neglected disease research, self-interest should nonetheless drive increased world support for indigenous research in African Universities. For 30 years, David Dunne has collaborated with excellent African human schistosomiasis researchers and argues that African research is not only vital for solving African problems, but for wider world health and knowledge. He is the Director of Cambridge-Africa, a vehicle for Cambridge's commitment to use the full extent of its expertise, facilities and influence to support African Universities across all academic disciplines (see: *http://www.thrive.cam.ac.uk*).

Wright Medal Lectures

West Road Auditorium 9:30am Tuesday 8th April

The British Society for Parasitology (BSP) awards an annual medal to commemorate the life of Dr Chris Wright, Natural History Museum, by formal recognition of an individual's research excellence and expertise in parasitology. The BSP 2014 Wright Medalists are Alex Rowe, University of Edinburgh, for her outstanding work on *Plasmodium* and Michael Barrett, University of Glasgow, for his huge contributions to trypanosomiasis research.

Modes of action and mechanisms of resistance to anti-protozoal drugs

Michael Barrett, University of Glasgow

My interest in Parasitology was triggered as an undergraduate Zoology student following a trip to Tanzania in 1985. I moved into PhD work in human African trypanosomiasis at the University of Cambridge, where I started work dissecting the biochemistry of the African trypanosome. I have continued in this field to this day, exploiting findings with the aim of learning how to target protozoa with new drugs and also learning about mechanisms of drug resistance. Most recently I have applied metabolomic analysis to learn more about drug action and I will summarise data revealing modes of action of various anti-protozoal agents and how resistance emerges.

Investigating a 100 year-old mystery: receptor-ligand interactions in severe malaria

Alex Rowe, University of Edinburgh

One of the hallmarks of fatal falciparum malaria is the presence of infected red cells clogging up small blood vessels in vital organs. This "sequestration" of infected cells was described in the 1890s, but the molecular mechanisms responsible for the adhesion of infected erythrocytes to human cells have only recently begun to be deciphered. Since starting my D.Phil. in Oxford in 1990, I have been studying the receptor-ligand interactions that lead to sequestration of malaria parasites, with the ultimate aim of developing adhesion-blocking therapies to treat or prevent severe malaria. What have we learned and what obstacles remain to translating these studies in basic science into clinically useful interventions?

Plenary Lectures

West Road Auditorium, 9am Monday 7th April and 9am Tuesday 8th April

The development of targeted anti-microbials to combat tuberculosis and the emergence of resistance

Sir Tom Blundell FRS, University of Cambridge

The spread of multidrug-resistant Mycobacterium tuberculosis poses important challenges to treatment with currently available anti-TB drugs and the situation is exacerbated by the emergence of extensively drug-resistant strains of M. tuberculosis. My talk will focus on current progress in using structure-guided fragment-based approaches to discover new drug candidates in collaborative research programmes funded by EU FP7 MM4TB and the Gates Foundation HIT-TB. I will also discuss computational approaches to identify targets of hits from phenotypic screening and to understand the basis of resistance described by recent second-generation gene sequencing.

Microparasites in space and time

Bryan Grenfell, Princeton University

Understanding the spatio-temporal spread of infectious diseases, and the impact of control measures, is an important research area for global health. Acute, immunizing viral infections provide a useful laboratory to study the determinants of spatiotemporal spread for directly-transmitted infections. We illustrate this with recent work on the spread of acute infections, focusing on measles, influenza, rotavirus and RSV, and the impact of control. We show that observed spatio-temporal patterns vary widely between infections, as a complex function of demographic and environmental drivers, herd immunity and viral evolution. Notwithstanding these intricacies, there are surprisingly strong signatures of multi-year predictability of epidemic dynamics in many settings. We conclude with a brief discussion of the general implications for spread and control of micro- and macroparasites.

The extreme evolutionary cell biology of trypanosomes

Keith Gull FRS, University of Oxford

The talk will provide an overview of the morphogenesis of the trypanosome cell and some fundamental and repeating principles are used in its construction. It will then explore how particular cell types within the life cycles of kinetoplastid parasites are fitted for their pathogenicity niches, and how transformation between the various forms is orchestrated. How such diversity of cellular form has evolved will be explored in the context of genome information and post-genomic technologies. Finally, an argument will be presented that the trypanosomes are superb examples of extreme cell biology **and are informative to the understanding of unity and diversity in evolutionary cell biology.**

Evolutionary cell biology of the Apicomplexa

David Roos, University of Pennsylvania

Recent advances in phylogenomic sampling and subcellular imaging provide new insights into the diversity and evolution of eukaryotic cells and organelles. Apicomplexan parasites harbor a nucleus, endosymbiotic structures (the mitochondrion, and even a plastid, acquired via secondary transfer from a eukaryotic alga), endomembrane system (ER, Golgi, regulated secretory granules), and cytoskeletal elements … but deploy these organelles in unusual ways. The phylum is defined by an 'apical complex' of specialized secretory organelles (micronemes, rhoptries) associated with host cell attachment, invasion, and establishment of the intracellular 'parasitophorous vacuole'. Cytoskeletal specializations include the 'inner membrane complex' responsible for assembling daughter parasites within the mother -- a process more akin to viral replication than binary fission. This distinctive process challenges traditional perspectives on eukaryotic cell cycle control, and allows apicomplexans to dispense with otherwise ubiquitous organelles (life without a lysosome!) As rapid proliferation is critical for parasite pathogenesis, understanding schizogony also offers prospects for therapeutic intervention

BSP Debate

West Road Auditorium, 9am Wednesday 9th April

Is vector control too damaging to be justified? *Yes* or *No?*

To abate parasitic diseases in medical, veterinary or wildlife settings there are a variety of options available but with any disease-specific intervention there are several perspectives and arguments both for an against a specific position. With regard to vector control of parasitic diseases, there is a balance between beneficial (i.e. human health) and detrimental (i.e. environmental perturbation) impact(s), and which also needs to be framed within economic considerations. For example, one method of vector control might be seen today, or in the future, as being desirable, but when applied, or is scaled-up, is found to be inappropriate or perhaps even counterproductive. *Is the strategy too costly for the predicted return?*

To engender discussion and also stimulate questions from BSP members and delegates we are fortunate to have assembled a team of experts in vector control and ecology, who will advance pro and con perspectives. These range from insect vector and snail intermediate host-borne diseases and will bring into discussion present and future genetic manipulation methods. There will be ample time to receive questions from the floor, taking an informal vote by a show of hands at the beginning and at the end to ascertain if opinions have been influenced or even changed.

Chair: **Professor Russell Stothard.**

Panel: **Professor Steve Torr** (University of Warwick, LSTM):
Tsetse/mosquito ecology and control.
Professor Sam Loker (University of New Mexico):
Snail/biological control and molecular genetics.
Dr Lynn Dicks (University of Cambridge):
Ecosystem analysis and pollinator conservation in farming.
Professor Martin Donnelly (LSTM):
Insect biology/evolution and insecticide resistance.

Stream co-ordinators and abstract selections

Many thanks to the stream co-ordinators and session chairs for their hard work in organizing an excellent programme and selecting abstracts for oral presentations. The following are specifically for their efforts with abstract selections;

Kinetoplastida (Trypanosome and Leishmania symposium)

Mark Carrington (University of Cambridge)

Apicomplexa

Julian Rayner (Sanger Institute)
Oliver Billker (Sanger Institute)

Helminths

David Dunne (University of Cambridge)

Ecology

Jo Cable (University of Cardiff)
Andy Fenton (University of Liverpool)
Rachel Norman (University of Stirling)
Olivier Restif (University of Cambridge)
Sarah Perkins (Cardiff University)

Veterinary Parasitology

Jacqui Matthews, Moredun Research Institute
Fiona Tomley, Royal Veterinary College
Damer Blake, Royal Veterinary College

Parasitology in the Middle East

Ashraf M. Ahmed (King Saud University)

Public understanding, outreach and communication

Tansy Hammarton (University of Glasgow)

Main Sessions

Day 1 - 07/04/2014

Session A1 (WRL) - Kinetoplastida - Genome structure and evolution

Chair: Steve Kelly, University of Oxford

11:00 AM - 11:30 AM (30 mins)
Slimming down and suiting up: key transitions in the evolution of trypanosomatids
Andrew Jackson
Department of Infection Biology Institute of Infection and Global Health University of Liverpool

A decade of comparative genomics has exposed the evolutionary changes that have shaped trypanosomatid genomes. Key transitions in the diversification of trypanosomatids, such as their origins from free-living phagotrophs, the evolution of an extracellular life strategy in the salivarian trypanosomes and the origin of *Leishmania* from trypanosomatids with a single insect host, can now be understood in terms of genomic reduction and innovation. African trypanosomes lack an intracellular life stage and have evolved antigenic variation as an adaptation for immune evasion. By comparing the variant antigen repertoires of salivarian trypanosomes, we have shown how these serve as reservoirs for genetic innovation, and how the process of phylogenetic turnover creates antigenic diversity in species-specific ways. *Leishmania* spp. have a two-host life cycle but originated from a single-host ancestor. Comparison of *Leishmania* spp. and closely related, monoxenic trypanosomatids has shown that innovation in gp46 and amastin genes accompanied this origin and continued to evolve during the differentiation of *Leishmania*. To explore the origin of all trypanosomatid parasitism, we have sequenced the genome of a free-living Kinetoplastid, *Bodo saltans*. The *B. saltans* gene repertoire provides the out-group to finally resolve long-standing issues such as the origins of key parasite adaptations, the mutual exclusivity of trypanosomatid cell surface architectures, the importance of horizontal gene transfer to the evolution of parasitism, and plesiomorphy versus genetic loss as explanations for trypanosomatid physiology. We will show how the trypanosomatid genome has been streamlined without losing function,

11:30 AM - 11:45 AM (15 mins)
Insights into 'missing biology' of *Leishmania mexicana* from RNA-sequencing
Eva Gluenz[1], Michael Fiebig[1], Steve Kelly[2], and Keith Gull[1]
[1.] Sir William Dunn School of Pathology, University of Oxford, South Parks Road, Oxford, OX1 3RE, UK; [2.] Department of Plant Sciences, University of Oxford, South Parks Road, Oxford, OX1 3RB, UK

Leishmania mexicana has two principal life cycle stages: promastigote forms in the sandfly and amastigote forms in mammalian phagocytes. Our aim was to define gene expression patterns characteristic of intracellular amastigotes and map transcript boundaries to enable further investigations into stage-specific biology and regulation of gene expression. We used Illumina sequencing of poly-A selected RNA to profile the transcriptome of promastigotes (PRO), axenic amastigotes (AXA) and amastigotes (AMA) in mouse bone marrow derived macrophages (BMDM). RNA-sequencing allows simultaneous sequencing of host and parasite RNA and we performed dual RNA-seq of the infected BMDMs. From the analysis of the *Leishmania* transcripts we determined global transcript abundance in PRO, AMA and AXA and tested for differential gene expression between cell types. We found 1665 of the annotated *L. mexicana* genes were differentially expressed between AMA and PRO; enrichment analysis identified cell surface proteins, transporters and peptidases among the transcripts more abundant in AMA.143 genes were differentially expressed between AMA and AXA and 576 between PRO and AXA, indicating that axenic amastigotes had an intermediate phenotype, sharing characteristics both with PRO and AMA. Mapping of splice and poly-A sites allowed us to generate the first detailed gene models for *L. mexicana*. We identified >1000 novel transcripts, the majority of which have the potential to encode small proteins (between 25 and 100 amino acids). Peptide evidence for several of these predicted proteins supports the idea of an as yet uncharacterised 'small proteome' in *Leishmania* with possible implications for host-pathogen interactions.

11:45 AM - 12:00 PM (15 mins)
Nuclear DNA Replication in *Trypanosoma brucei*: the beginning *

Catarina A. Marques, Nicholas J. Dickens, Calvin Tiengwe, Lucio Marcello, and Richard McCulloch
Wellcome Trust Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, University of Glasgow

Little is known about how DNA replication takes place in *Trypanosoma brucei*. Presently, studies strongly suggest that the molecular machinery involved in the recognition of replication starting sites in *T. bruce*i might be considerably different from the one typically found in eukaryotes – the six-subunit (Orc1-6) origin recognition complex (ORC). For instance, several key players involved in the initiation of DNA replication in eukaryotes are not found in *T. brucei*, and only five ORC-like factors have been identified: three apparent distant orthologues of ORC subunits (TbORC1/CDC6, TbORC1B, and TbORC4), and two kinetoplastid-specific factors, Tb7890 and Tb3120. These factors have been shown to interact with TbORC1/CDC6, but whether they form a complex has not been shown. We aim to elucidate how DNA replication is initiated in *T. brucei*, specifically whether these factors act as a divergent ORC-like complex. Currently, our data on the localisation of TbORC1/CDC6, TbORC4 and Tb3120 (nuclear throughout the cell cycle) support this idea, while individual silencing of these factors by RNA interference results in a impairment of DNA replication, confirming their involvement in this process. Interestingly, data indicate that TbORC1B is essential for DNA replication, and together with its restricted localisation to the nucleus of S-phase cells, strongly suggests that TbORC1B might be a regulatory factor rather than a static member of a complex. S-phase restriction of a putative ORC-like factor may indicate pronounced divergence in the machinery and regulation of DNA replication in *T. brucei*, which might be used as a potential drug target against trypanosomes.

12:00 PM - 12:15 PM (15 mins)

The genome of *Trypanosoma rangeli*, a trypanosomatid avirulent to mammals

Edmundo Grisard, and Patrícia Hermes Stoco
Universidade Federal de Santa Catarina, Florianópolis, SC, Brazil, 88040-970

Trypanosoma rangeli infects humans as well as wild and domestic mammals across Central and South America. It is non-pathogenic to mammals but can be mistaken for the etiologic agent of Chagas disease (*T. cruzi*). The sequencing of the *T. rangeli* genome revealed a haploid genome of ~24Mb in length, being the smallest and less repetitive trypanosomatid genome obtained. This parasite genome has shorter subtelomeric sequences compared to those of *T. cruzi* and *T. brucei*, displays intraspecific karyotype variability and lacks minichromosomes. Among the 7,613 coding sequences predicted, the functional annotation of 2,415 were validated, while 4,999 were hypothetical proteins, and due the support of EST and MS, 44 were proteins of unknown function. A DCL 2 gene ortholog involved on the *T. brucei* RNAi pathway was found in *T. rangeli*, but the RNAi machinery is non-functional since the other elements are present as pseudogenes. *T. rangeli* is highly susceptible to oxidative stress, a phenotype that may be explained by a smaller number of anti-oxidant defence enzymes and heat-shock proteins. Phylogenetic comparison of nuclear and mitochondrial genes indicates that this species diverged from the *T. cruzi* lineage after separation from the African salivarian trypanosomes. In addition of revealing new aspects of trypanosome co-evolution within the vertebrate and invertebrate hosts, comparative genome analysis with pathogenic trypanosomatids provides new tools for elucidating aspects of the biology of this species and key pathways related to host-trypanosome interactions aiming the development of diagnostic tools and/or therapeutic targets. Supported by CAPES and CNPq.

12:15 PM - 12:30 PM (15 mins)

Aneuploidy in natural *Leishmania* populations: chromosome chaos or adaptive strategy?

Jean-Claude Dujardin[1], Imamura H[1], Mannaert A[1], Rijal S[2], Sundar S[3], Arevalo J[4], Bhattarai N[2], Tihon E[1], Van Den Abbeele J[1], Carter K[5], Gamarro F[6], Berriman M[7], and Cotton J[7].

[1.] Institute of Tropical Medicine, Antwerp; [2.] BPKIHS, Dharan, Nepal; [3.] BHU, Varanasi, India; [4.] IMTAvH, Lima, Peru [5.] Strathclyde University, Glasgow, UK; [6] IPBLN-CSIC, Granada, Spain [7.] WTSI, Hinxton, UK

Aneuploidy has been extensively demonstrated in *Leishmania* under experimental conditions (stress, drug pressure…), but the frequency and the significance of this phenomenon in natural populations of trypanosomatids is yet to be defined. In the frame of three running projects on whole genome sequence diversity, we explored the occurrence of aneuploidy among clinical isolates of *L. donovani* (203 lines sequenced, Indian sub-continent), *L. braziliensis* (53 lines, Peru and Bolivia) and *Trypanosoma congolense* (45 lines, sub-Saharan Africa). In *T. congolense*, deviations from disomy were not encountered in any of the 11 chromosomes. In sharp contrast, aneuploidy is frequent in the 2 *Leishmania* species: (i) chromosome 31 was tetrasomic in all strains and (ii) 33 and 26 chromosomes showed a variable ploidy in *L. donovani* and *L. braziliensis* respectively. However, some near-diploid isolates (all disomic chromosomes, except for number 31) are observed: 20 and 30% in both species respectively. Analysis of ploidy patterns over a decade of sampling shows some changes between near-diploid and aneuploid parasites, with different aneuploid chromosomes recurrently observed in each species: chr 8, 9, 20, 23 and 33 in *L. donovani* and chr 11 and 25 in *L. braziliensis*. Our data do not support the presence of less repeated genes in chromosomes as a major driving force for aneuploidy. Aneuploidy is likely an adaptive strategy, driven by selective pressure on the chromosomal gene content, as shown in experimentally induced drug resistance.

Session B1 (WRR) - Apicomplexa Cell Biology

Chair: Julian Rayner, Wellcome Trust Sanger Institute

11:00 AM - 11:30 AM (30 mins)
Single Molecular, Structural and Biochemical insights into actin regulation in the malaria parasite
Jacob Baum
Imperial College London

Malaria parasites cycle between two hosts during their complex lifecycle: the mosquito and human. Each lifecycle stage is adapted to its specific target tissue, yet retains a conserved way of moving, called gliding, based on an actin-myosin motor. Gliding motility is an ancient, yet seemingly simple, way of crossing substrates and entering host cells and is central to understanding the evolution of substrate-dependent eukaryotic cell motility. Its unique nature also makes the gliding motor an attractive target for new antimalarial drugs. However, little is known about the dynamics of the cytoskeletal machinery at its core, in particular the behaviour of malaria actin – the most divergent among eukaryotes – and central regulator of active motion in the parasite. Work in the Baum laboratory has been investigating the biochemical and structural properties of the core actin regulating proteins in the malaria parasite towards understanding how motility itself might be controlled. Malaria parasites contain a remarkably reduced repertoire of actin regulatory proteins when compared to other eukaryotic cells making it a surprisingly minimal actin system to study. In the process of analysing some of these key regulators we have revealed several fundamental insights into not only parasite actin regulation but also its control across eukaryotes; the black sheep teaching lessons across the family. Here I will present an update of recent structural, biochemical and single-molecule imaging insights into how actin and motility in general might be controlled in the parasite cell.

11:30 AM - 11:45 AM (15 mins)
Adhesion properties of *P. falciparum* infected erythrocytes that bind human brain endothelial cells *
Yvonne Azasi and J. Alexandra Rowe
Centre for Immunity, Infection and Evolution, Institute of Immunology and Infection Research, School of Biological Sciences, University of Edinburgh, Edinburgh, EH9 3JT, United Kingdom

Cerebral malaria is characterized by sequestration of mature *Plasmodium falciparum* infected erythrocytes (IEs) in microvasculature of the brain. The IEs sequester to avoid splenic clearance resulting in impaired perfusion and death of the host. Variant surface antigen, *Plasmodium falciparum* erythrocyte membrane protein one, encoded by var genes of domain cassettes (DC) 8 and 13, have been identified as the parasite ligands for cytoadherence of IEs to brain endothelial cells. Endothelial Protein C Receptor (EPCR) has been identified as the host receptor for adhesion. Knowledge of interactions involved in the adhesion process is essential for development of adjunctive therapies to reverse cytoadherence in cerebral malaria. Using human brain endothelial cell line, HBEC-5i and three (DC8 or DC13-expressing) parasite lines, we investigated effect of pH, temperature, parasitaemia, gaseous conditions and serum on adhesion to HBEC-5i, and the role of six endothelial cell receptors (EPCR, CD36, ICAM1, PECAM1, CSA and heparin) in adhesion. Cytoadhesion was found to increase with parasitaemia, maximum at pH 7.2 to 7.3, and significantly inhibited by human serum in all strains. Room temperature versus 37oC and 39oC, and hypoxic versus normoxic conditions did not significantly affect adhesion. EPCR was confirmed to be a receptor for binding of IT4var19-expressing parasites. However, EPCR antibodies and recombinant protein did not inhibit adhesion of HB3var3- and IT4var7-expressing parasites. This study helps elucidates optimal conditions for adhesion of IEs to brain endothelial cells. It also suggests that EPCR is not the only receptor involved in cytoadherence of DC8 and DC13-expressing IEs in the brain.

11:45 AM - 12:00 PM (15 mins)

Exploring the membrane-microtubule interaction at the cortex of *Toxoplasma gondii*

Ke Hu[1], Jun Liu[1], Laura Wetzel[1,2], Ying Zhang[3], Eiji Nagayasu[4], Stephanie Ems-McClung[5], Laurence Florens[3]

[1.]Department of Biology, Indiana University, Bloomington, IN, 47405, USA [2.]Current address: Department of Molecular and Cellular Biology, University of California, Berkeley, CA, 94720, USA [3.]Stowers Institute for Medical Research, Kansas City, MO, 64110, USA [4.]Department of Infectious Diseases, Division of Parasitology, Faculty of Medicine, University of Miyazaki, Miyazaki, Japan [5.]Medical Science Program, Indiana University, Bloomington, IN 47405, USA

Toxoplasma gondii has several distinct tubulin-containing structures, including 22 cortical microtubules. These microtubules are required for the replication and for maintaining the shape of the parasite. They are extremely stable and form extensive interactions with the membrane pellicle, properties important for the essential functions of these microtubules. These features are not found in the microtubules of the host, therefore are potential drug targets. Early ultrastructural studies have shown that the cortical microtubules are heavily decorated with associated proteins, which are likely responsible for the novel properties of these polymers. However, only two of these proteins were known and they cannot explain the essential function of the cortical microtubules. We have recently identified the first protein complex associated with the cortical microtubules, which we named CMAC (for cortical microtubule associated complex). Six new novel proteins were found in this complex, three of which are conserved between *T. gondii* and *Plasmodium falciparum*. Super-resolution Structured-illumination imaging of parasites expressing fluorescently-tagged CMAC components produced a detailed view of the microtubule cytoskeleton. Interestingly, some of these proteins only decorate a segment of the cortical microtubules, which might be correlated with the segmentation of the membrane pellicle revealed by previous EM studies. Preliminary evidence shows that CMAC components play an important role in regulating the organization of the cortical microtubules and the parasite shape. Further study of this set of novel proteins will provide insights into how the biochemical properties of the protein coat on the cortical microtubules dictate the organization of the membrane pellicle.

12:00 PM - 12:15 PM (15 mins)

Processing of *Plasmodium falciparum* merozoite surface protein 1 is essential for blood stage parasite viability. *

Sujaan Das, Christine R Collins, Nadine Hertrich, Dominique Soldati-Favre, Christian Epp, Michael J Blackman

MRC National Institute for Medical Research, University of Heidelberg, and University of Geneva

The *Plasmodium* merozoite is covered by an essential GPI-anchored protein called Merozoite-Surface-Protein-1 (MSP1). MSP1 is synthesised as a precursor that is proteolytically processed prior to egress by the parasite subtilisin-like protease 1 (SUB1) to yield 4 polypeptides of ~83, 30, 38 and 42kDa. These remain bound to the merozoite surface until being shed at invasion by a distinct protease, SUB2. Despite years of research, the function of MSP1 and of its proteolytic cleavage remains largely unknown. We hypothesise that processing of MSP1 is important for its function and hence parasite survival. Using targeted mutagenesis of the *P. falciparum* msp1 locus and knowledge of SUB1 substrate-specificity, we have investigated the effects of mutations that block SUB1-mediated cleavage of MSP1. We show that processing at the membrane-proximal 38/42 cleavage-site in fact occurs at three closely spaced redundant positions, and that at least one cleavage is necessary and sufficient for parasite survival. Point mutations that block individual cleavage-sites, or two simultaneously, are tolerated and shift cleavage to the next available cleavage-site. Mutations or deletions that simultaneously block cleavage at all three sites are lethal. The absolute requirement for cleavage in the 38/42 region of MSP1 provides the first definitive evidence that SUB1-mediated processing of MSP1 is essential for MSP1 function. Our recent data using recombinant MSP1 and binding assays have indicated that processing of MSP1 acts to modulate its binding affinity for an erythrocyte surface receptor. This finding is the first to link protease-mediated processing to the function of a malaria merozoite surface protein.

12:15 PM - 12:30 PM (15 mins)

Phosphoinositide metabolism links cGMP-dependent protein kinase G to essential Ca^{2+} signals at key decision points in the life cycle of malaria parasites

Mathieu Brochet[1], Mark O Collins[1,2], Terry K Smith[3], Eloise Thompson[4], Sarah Sebastian[1], Katrin Volkmann[1], Frank Schwach[1], Lia Chappell[1], Ana Rita Gomes[1], Matthew Berriman[1], Julian C Rayner[1], David A Baker[4], Jyoti Choudhary[1] and Oliver Billker[1]

[1]Wellcome Trust Sanger Institute, Hinxton, Cambridge CB10 1SA, UK [2]Present address: Department of Biomedical Science, Firth Court, Western Bank, Sheffield, S10 2TN, UK [3]Schools of Biology & Chemistry, Biomedical Sciences Research Complex, The North Haugh, The University of Saint Andrews, KY16 9ST, UK. [4]Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT, UK

Many critical events in the *Plasmodium* life cycle rely on the controlled release of Ca^{2+} from intracellular stores to activate stage-specific Ca^{2+}-dependent protein kinases. Using the motility of *P. berghei* ookinetes as a signalling paradigm we show that the cyclic guanosine monophosphate (cGMP)-dependent protein kinase, PKG, maintains the elevated level of cytosolic Ca^{2+} required for gliding motility. We find that the same PKG-dependent pathway operates upstream of the Ca^{2+} signals that mediate activation of *P. berghei* gametocytes in the mosquito and egress of *P. falciparum* merozoites from infected human erythrocytes. Perturbations of PKG signalling in gliding ookinetes has a marked impact on the phosphoproteome, with a significant enrichment of in vivo regulated sites in multiple pathways including vesicular trafficking and phosphoinositide metabolism. A global analysis of cellular phospholipids demonstrates that in gliding ookinetes PKG controls phosphoinositide biosynthesis, possibly through the subcellular localisation or activity of lipid kinases. Similarly, phosphoinositide metabolism links PKG to egress of *P. falciparum* merozoites, where inhibition of PKG blocks hydrolysis of phosphatidylinostitol (4,5)-bisphosphate. In the face of an increasing complexity of signalling through multiple Ca^{2+} effectors, PKG emerges as unifying factor to control multiple cellular Ca^{2+} signals essential for malaria parasite development and transmission.

Session C1 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment I

Chair: Alan Fairlamb, University of Dundee

11:00 AM - 11:30 AM (30 mins)
Gene expression in trypanosomes – the control freak's guide
Isabela Ribeiro
Institute of Cell Biology, University of Bern, Bern, Switzerland.

Post-transcriptional regulation, in particular RNA stability, has long been regarded as the central mechanism for regulating gene expression by trypanosomes. It has been known for some time that the parasites employ a much larger set of control mechanisms to regulate expression of the major surface proteins, which are transcribed by RNA polymerase I. These include regulation of transcription initiation, elongation, RNA maturation and translation. Recent genome-wide analyses indicate that multiple layers of regulation are the rule, not the exception, and that chromatin structure and co-transcriptional control mechanisms also contribute to determining expression levels of messenger RNAs from the polycistronic clusters transcribed by RNA polymerase II. At the next level, cohorts of transcripts, whose genes are dispersed throughout the genome, can form regulons whose RNA stability or translation are coordinately regulated. Relatively few regulatory proteins have been identified to date. Genome-wide RNAi screens and comparative RNA-Seq have recently led to the identification of novel regulators of stage-specific expression. These include conserved housekeeping proteins that seem to have acquired additional functions in trypanosomes.

11:30 AM - 11:45 AM (15 mins)
Biochemical characterization and *in vivo* chemical validation of *Trypanosoma brucei* phosphodiesterases, as potential drug target
Srinivasa P S Rao, Christian Noble, Chin Chin Lim, Kelly Chew, Rishi Arora, Pamela Thayalan, Vanessa Manoharan, Fiona Ng, Cyrille Kounde, Ida Ma, Kirk Wright, Prakash Vachaspati, Peter Gedeck, Timothy Benson, Wan Kah Fei, Manjunatha Ujjini, and Pearly Ng
Novartis Institute for Tropical Diseases, #05-01, Chromos, 10 Biopolis Raod Singapore 138670

The current anti-trypanosomal therapies suffer from problems of toxicity and inadequate efficacy hence there is an urgent need for safer and efficacious drugs. Phosphodiesterases (PDE) are promising druggable targets which are highly specific hydrolases that convert the signaling molecule cyclic adenosine (cAMP) into AMP. *Trypanosoma brucei* (Tb) has five PDE homologs, of which PDEB1 and B2 are essential for survival of blood-stage form. Although, the catalytic domain of TbPDE's are highly conserved to human PDE4, it has unique 'parasite pocket' adjoining to the active site which could be exploited for selectivity. In this study, we report the biochemical characterization, crystallization and in vivo chemical validation of TbPDE, as potential drug target. Recombinant expression and purification of TbPDEB1 and B2 using the baculovirus expression system yielded cAMP-specific TbPDE enzymes. Multiple known PDE inhibitors such as Piclamilast and a tool compound inhibited TbPDE with IC50 concentrations of 9.9 and 0.15mM. The tool compound inhibited Tb growth both in vitro and in vivo in a blood-stage mouse-efficacy model. We have obtained a high-resolution (1.88Å) crystal structure of TbPDEB2. We have used multiple screening approaches to identify novel PDE inhibitors such as medium throughput screening of focused libraries using the TbPDE enzyme; structure-based drug design (SBDD) and fragment-based screening (FBS). Co-crystallization of compounds obtained by SBDD and FBS will be useful in guiding medicinal chemistry efforts for identification of TbPDE-specific lead molecules. Thus in vivo chemical validation of target and new lead discovery efforts will potentially lead to promising novel Tb-specific PDE inhibitors.

11:45 AM - 12:00 PM (15 mins)
Targeting the nucleotide metabolism of *Trypanosoma brucei* I
Anders Hofer, Farahnaz Ranjbarian, and Munender Vodnala
Dept. of Medical Biochemistry & Biophysics, Umeå University, Sweden

The current chemotherapy against African sleeping sickness is limited by toxicity or low efficacy against all variants/stages of the disease. Nucleoside analogs are interesting as possible drug candidates since many of them are

known to pass the blood brain barrier, which is a prerequisite to cure late-stage African sleeping sickness. We have characterized enzymes involved in nucleotide metabolism of the parasite and its human host in order to develop nucleoside analogs that specifically target *T. brucei*. The parasite has two nucleoside kinases; adenosine kinase and thymidine kinase, which phosphorylate the analogs to their nucleotide forms and subsequently inhibit nucleotide-depending reactions such as RNA and DNA synthesis. Interestingly, the phosphorylation of many adenosine analogs also leads to ATP depletion in the parasites since their highly efficient adenosine kinase consumes most of the cellular ATP when it phosphorylates the analogs. When developing nucleoside analogs against the parasite, it is also important to consider nucleoside degradation pathways. Of particular importance is methylthioadenosine phosphorylase which protects *T. brucei* from deoxyadenosine toxicity by cleaving this nucleoside into adenine and deoxyribose-1-phosphate. We have found that that the protection system can be circumvented by cleavage-resistant substrate analogs of adenosine kinase. Some of these analogs are 1000-fold more active on *T. brucei* than mammalian cells and cure *T. brucei*-infected mice without any obvious side effects.

12:00 PM - 12:15 PM (15 mins)

Inhibitors of *Leishmania major* Inositol Phosphorylceramide Synthase - New Therapies for Leishmaniasis

Christopher Brown[1], John G. M. Mina[1], Andy T. Merritt[2], Paul W. Denny[1], and Patrick G. Steel[1]
[1]Biophysical Sciences Institute, Durham University, Durham DH1 3LE, [2]Medical Research Council Technology, Mill Hill, London NW7 1AD

Leishmaniasis is a Neglected Tropical Disease (NTD) caused by the protozoan *Leishmania* spp. Over 350 million people worldwide are at risk, with an estimated annual death toll of 60,000. Treatment typically requires long, expensive courses of exposure to toxic medicines via parenteral administration. There is an urgent need for novel treatments that are inexpensive and free of side effects. Our group has previously identified and validated the *Leishmania major* inositol phosphorylceramide synthase (LmjIPCS) enzyme as an attractive drug target(1,2). This essential membrane-bound enzyme has no mammalian equivalent, potentiating the development of safe, selective anti-leishmanials. A set of 1200 pharmacologically active compounds (NINDS set supplied by MRCT) were screened against LmjIPCS, 57 were found to exhibit >70% inhibition at 20 µM. A secondary assay against *L. major* promastigotes highlighted 14 compounds with selective cytotoxic effects. Four of these inhibitors displayed ED50 values lower than that of pentamidine (2.05 µM), a second-line treatment for leishmaniasis. Significantly these compounds have good bioavailability and mammalian safety profiles, making them ideal for development into new drugs. A hit-to-lead study is being explored to elucidate further structure-activity relationship data. This approach has the scope to provide a wide variety of chemical tools that will deliver potent, selective leishmanicidal therapies. References: (1) P.W. Denny, H. Shams-Eldin, H.P. Price, D.F. Smith, R.T. Schwarz, J. Biol. Chem., 2006, 281, 28200- 28209; (2) J.G. Mina, J.A. Mosely, H.Z. Ali, H. Shams-Eldin, R.T. Schwarz, P.G. Steel, P.W. Denny, Int. J. Biochem. Cell Biol., 2010, 42, 1553-1561.

12:15 PM - 12:30 PM (15 mins)

A robust *in vivo* imaging model for late stage human African trypanosomiasis to evaluate anti-trypanosomal drugs. *

Hollie Burrell-Saward, Vanessa Yardley, Martin Taylor, John Kelly, Simon Croft, Theresa Ward.
ITD London School of Hygiene & Tropical Medicine Keppel Street London WC1E 7HT

There are 10,000 reported cases of human African Trypanosomiasis (HAT) annually, with 60 million people at risk. Despite this, HAT is a Neglected Tropical Disease with few significant advances in chemotherapy over the last 50 years. Limited knowledge of the metabolic status and drug sensitivity of trypanosomes in the central nervous system (CNS), and the lack of a useful murine CNS model has limited drug discovery. Here, we report the generation of highly bioluminescent parasites and their use in an in vivo imaging model of stage II African trypanosomiasis. Bloodstream forms of the chronic model strain GVR35 were transformed with a construct designed to express “red-shifted” luciferase. Using the standard 21 day treatment model in CD1 mice, we were able to identify CNS infection after treatment with a stage I drug (Berenil). By using the bioluminescence model in a drug relapse experiment with melarsoprol, early relapse could be indentified before peripheral blood parasitaemia could be detected. This provides evidence that the model can be used as to reduce the current 180-day experiment. Trypanosome numbers quantified by qPCR correlated with luciferase intensity.

Session D1 (RCA) - Helminths- Diagnostics & Control

Chair: Joanne Webster, Imperial College London

11:00 AM - 11:30 AM (30 mins)

Diagnosing 'Egg-negative' Schistosomiasis, a Modern Priority in the Move toward Elimination

Charles King

Center for Global Health and Diseases, Case Western Reserve University, Cleveland, Ohio USA and Schistosomiasis Consortium for Operational Research and Elimination (SCORE, University of Georgia Athens, Georgia USA

The reduction of *Schistosoma* prevalence in many countries by means of mass drug administration has been highly encouraging, but unmasks the fact that drugs alone cannot prevent reinfection or eliminate the risk of infection-associated disability. More sensitive means for diagnosis of *Schistosoma* infection have become essential for defining: i) the true disease burden of infection, ii) the treatment efficacy of established or experimental therapeutic s, and iii) the protection afforded by candidate vaccines. Attempts at local elimination, now sanctioned by the World Health Assembly WHA 66.12, will require effective means of detecting low-intensity infections, which can often prove 'egg-negative' on standard stool or urine parasitology. Recent progress in the stable detection of circulating parasite antigens, of short-lived anti-parasite antibody isotypes, and of parasite DNA all suggest means to successfully monitor ongoing parasite transmission and persisting human infection as community prevalences approach nil.

11:30 AM - 11:45 AM (15 mins)

Schistosomiasis elimination in Zanzibar (Unguja and Pemba Islands): design and implementation of an integrated multidisciplinary research programme

Stefanie Knopp[1,2,3], Khalfan A. Mohammed[4], Said A. Mohammed[5], Bobbie Person[6], Shaali M. Ame[5], David Rollinson[1]

[1]Department of Life Sciences, Natural History Museum, London, United Kingdom [2]Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, Basel, Switzerland [3]University of Basel, Basel, Switzerland [4]Helminth Control Laboratory Unguja, Ministry of Health, Zanzibar, United Republic of Tanzania [5]Public Health Laboratory - Ivo de Carneri, Pemba, United Republic of Tanzania [6]Independent Consultant, Schistosomiasis Consortium for Operational Research and Evaluation, University of Georgia, Athens, United States of America

Gaining and sustaining control of schistosomiasis and, whenever feasible, achieving local elimination are the year 2020 targets set by the World Health Organization. In Zanzibar, various institutions and stakeholders have joined forces to eliminate urogenital schistosomiasis transmission within 5 years. In a randomized intervention trial, we aim to compare the impact of biannual mass drug administration (MDA) of praziquantel to the whole at-risk population (arm 1), MDA plus snail control interventions (arm 2), and MDA plus behaviour change interventions (arm 3) in a total of 45 communities each on Unguja and Pemba island. Since the onset of the project in November 2011, four praziquantel treatment rounds have been conducted with a reported coverage of around 80%. Snail control started in August 2012 and almost 100 natural freshwater bodies in 30 communities are treated regularly with niclosamide when intermediate host snails (*Bulinus globosus*) are present. Behaviour change interventions were designed together with the communities in additional 30 communities. Implementation of urinals, teacher's packages, safe play for children, and laundry areas commenced in October 2012. The first parasitological follow-up survey conducted in early 2013 did not reveal an impact of any intervention. Prevalences and infection intensities decreased in some communities but increased in others compared with baseline. *Bulinus globosus* returned into most of the treated water bodies. In the behavioural study communities, students' knowledge about schistosomiasis transmission and prevention increased. Challenges for the research programme include sample size, infection hot-spots, migration, and suboptimal adherence to drug intake.

11:45 AM - 12:00 PM (15 mins)

The insulin receptor: an Achilles' heel for schistosome vaccine development

Donald McManus[1], Hong You[1], Geoffrey N. Gobert[1], Mary G. Duke[1], and Rachel J. Stephenson[2],

[1.]Molecular Parasitology Laboratory, Infectious Diseases Division, QIMR Berghofer Medical Research Institute, Brisbane, Queensland, Australia. [2.]Institute of Molecular Biosciences, University of Queensland, Brisbane, Queensland, Australia

Schistosomes consume considerable amounts of glucose but depend on host insulin for its uptake. We isolated two human insulin receptors (SjIRs) from Asiatic *Schistosoma japonicum* which bind human insulin in a manner similar to that of mammalian cells. We have identified the major insulin binding sites of the SjIRs using synthetic peptides and protein interaction and predicted structure assays.We demonstrated in mice that immunization with the L1 subdomain (insulin

binding domain) of the recombinant SjIR proteins conferred highly significant reductions in fecal eggs, stunting of adult worms, a reduction in liver granuloma density and a reduction in the numbers of mature intestinal eggs. Further, we found these molecules depressed long-term female growth and egg production in *S. japonicum* in mice, re-emphasizing their vaccine potential. Finally, when we knocked down the SjIRs using dsRNA in adult *S. japonicum in vitro*, we found their expression levels and those of other important downstream proteins in the insulin pathway decreased significantly, resulting in depressed development and growth of adult worms. These data underpin our efforts to develop a veterinary transmission blocking vaccine against *S. japonicum* as bovines are major reservoir hosts, being responsible for up to 90% of environmental egg contamination in China. We found the peptides derived from the SjIRs are conserved in schistosomes, suggesting all species infecting humans share the same motifs for insulin binding, so that targeting the IR could be readily extended to the African schistosomes, *S. mansoni* and *S. haematobium*, leading to human vaccine trials against both parasites.

12:00 PM - 12:15 PM (15 mins)

Schistosomiasis in pre-school-aged children and their mothers in Chikhwawa district, southern Malawi with notes on the local freshwater snail fauna

Russell Stothard, H. Poole, D.J. Terlouw, D. Lalloo
Department of Parasitology Liverpool School of Tropical Medicine Pembroke Place Liverpool L3 5QA

In June 2012, a parasitological survey of pre-school-aged children (PSAC) and their mothers assessed the extent of schistosomiasis in Chikhwawa district, Malawi. Diagnosis was made by host serology, supplemented by urine-antigen testing and a questionnaire-interview with mothers. Potential urinary tract morbidity was estimated by direct quantification of haematuria and albuminuria. In total, 49.5% (CI95 42.6-56.4) of 208 PSAC and 94.5% (CI95 90.9-98.1) of 165 mothers were seropositive for schistosomiasis by detection of antibodies against soluble egg antigen. Analysis of information obtained by questionnaire inferred extensive daily water contact of PSAC, with many having haematuria and albuminuria (~25%), indicative of active urinary tract lesions. Egg-patent infections of both urogenital and intestinal schistosomiasis were found and a selection of schistosome eggs were characterised by DNA barcoding. Concurrently, a malacological survey of freshwater habitats was made to investigate the potential for local transmission. Although Biomphalaria was not found, snail surveys encountered several populations of Bulinus africanus group species. Presently overlooked in terms of their disease and presently not included for treatment within the Malawian National Control Programme, PSAC have a high burden of disease and therefore require immediate access to praziquantel treatment.

12:15 PM - 12:30 PM (15 mins)

A large-scale school based deworming programme in Bihar State, India - recipe for success.

Laura Appleby, Prerna Makka, Sarman Singh, C.K. Mishra, Sanjay Kumar, Rajesh Bhushan, Rakesh Kumar, Kriti Sharma, Yogita Kumar, Late Sri Raman, Stalin Chakrabarty, Jimmy H Kihara, Ruth Dixon, Sanjay Kumar, and Lesley Drake
Deworm the World, Washington DC, USA; Partnership for Child Development, Imperial College, London, UK; All India Institute of Medical Sciences, New Delhi, India; Department of Health and Family Welfare, Government of Bihar, India; State Health Society, Bihar, India; Bihar Education Project Council, Bihar, India; Eastern and Southern Africa Centre of International Parasite Control, Kenya Medical Research Institute, Nairobi, Kenya

Soil-transmitted helminths (STH) are among the most common chronic infections and affect over 600 million school aged children worldwide, leading to physical, nutritional and cognitive impairment during crucial stages of growth and development. Drug delivery using a school based platform provides an efficient mechanism for distributing deworming tablets in endemic environments and has been shown to reduce school absenteeism and increase the potential to learn while still at school. Preliminary surveys conducted in Bihar State, India indicated prevalence levels of between 49% and 80% for any STH infection across the state, thus warranting mass drug administration (MDA) for all children twice a year. Between February and April 2011, 17 million school-aged children in India's Bihar State were dewormed as part of one of the largest school-based deworming efforts ever conducted in the world. The state-wide programme involved close partnership between health and education sectors and has ensured sustainability for ongoing annual deworming activities across Bihar. The success of this large-scale deworming programme is attributed to effective use of school based platforms, enabling environments consisting of coordinated efforts between policy stakeholders, partner organizations, community mobilization and team members on the ground. The programme provides a model that can be rapidly scaled up in other States and sustained overtime to improve education, health and productivity of school children. The background behind the implementation of this successful deworming programme, the conditions created to allow for its sustainability, as well as the challenges overcome within this large scale deworming programme will be discussed.

Session E1 (RCU) - Ecology - Bridging scales

Chair: Andy Fenton, University of Liverpool

11:00 AM - 11:30 AM (30 mins)
Should we link within- and between-host levels in evolutionary epidemiology?
Samuel Alizon
CNRS

In epidemiology, infected hosts are often treated as "black boxes" in which within-host dynamics are summarised with a few parameters such as transmission rate or infection duration. Over the last decade, nested models have been developed that incorporate within-host dynamics into evolutionary epidemiology settings. Is this extra complexity worth the effort? Indeed, too often the addition of the within-host level only results in a couple of extra more or less realistic parameters in the epidemiological model. However, in some cases, e.g. multiple infections, the nesting can prove to be essential. Bridging scales is facilitated by new tools that help incorporate within-host times series into evolutionary epidemiology models but there are still open challenges, especially to study rapidly evolving parasites. As illustrated by HIV genomic evolution data, most of the difficulties reside in the transmission bottleneck and in the fact that what gets in the host can differ from what gets out, i.e. that infection traits are not necessarily heritable from one infection to the next.

11:30 AM - 11:45 AM (15 mins)
Measuring health for parasitologists: Global Burden of Disease and beyond? *
Thomas Fürst, Lesong Conteh Maria-Gloria Basáñez
Centre for Health Policy, Imperial College London Department of Infectious Disease Epidemiology, Imperial College London

Over the past years, and particularly in the wake of the Global Burden of Disease studies, the quantification of health by means of the so-called health-adjusted life years (HALYs) has increasingly gained attention. Particularly, quality-adjusted life years (QALYs) and disability-adjusted life years (DALYs) have risen to prominence. These measures combine crude numbers of infected and cases of death by using weights to adjust for individuals' life-years spent in less than perfect health. An explicit assumption is that such quality- or disability-adjusted summary measures allow for direct comparison of different causes of health losses and potential mitigation strategies across populations. Not surprisingly, therefore, QALYs and DALYs are frequently used as denominators in cost-effectiveness analyses and have become a powerful "currency" in public health. Consequentially, it may become increasingly important for monitoring and evaluation of parasitic infection control programmes to consider not only the classical incidence, prevalence, morbidity and mortality indicators, but also broader notions of health-related quality of life and disability. We reviewed the literature and—largely based on the highly topical International Classification of Functioning, Disability and Health of the World Health Organization—developed a conceptual framework to delineate the concepts of health-related quality of life and disability. The presented conceptual framework may help to assess more comprehensively and systematically the negative impacts of parasitic infections and the benefits of their control. Furthermore, our considerations can act as a timely guide to understand better what recent QALY and DALY estimates do, and do not, encompass.

11:45 AM - 12:00 PM (15 mins)
Epidemiological feedbacks affect evolutionary emergence of pathogens
Matthew Hartfield, and Samuel Alizon
Laboratoire MIVEGEC (UMR CNRS 5290, IRD 224, UM1, UM2), 911 Avenue Agropolis, B.P. 64501, 34394 Montpellier Cedex 5, France

The evolutionary emergence of new pathogens via mutation poses a considerable risk to human and animal populations. Most previous studies have investigated cases where a potentially pandemic strain emerges though mutation from an initial, maladapted strain (that is, its basic reproductive ratio $R0 < 1$). However, an alternative (and arguably more likely) cause of novel pathogen emergence is where a 'weakly-adapted' strain (with $R0 \approx 1$) mutates into a strongly-adapted strain (with $R0 \gg 1$). In this case, a proportion of the host susceptible population is removed as the first strain spreads, but the impact this feedback has on emergence of mutated strains has yet to be quantified. We produce a model of

pathogen emergence that takes into account changes in the susceptible population over time, and find that the ongoing depletion of susceptible individuals by the first strain has a drastic effect on the emergence probability of the mutated strain, above that assumed by just scaling the reproductive ratio. Finally, we apply our model to the possible emergence of chikungunya virus on La Réunion island, and demonstrate that the emergence probability of the mutated strain was reduced approximately 10-fold, compared to models assuming that susceptible depletion would not affect outbreak probability. These results highlight the importance of taking population feedbacks into account when predicting disease emergence.

12:00 PM - 12:15 PM (15 mins)

Do patterns of nestedness in parasite communities of wild wood mice predict order of infection in individual hosts?

Evelyn C. Rynkiewicz[1], Andy Fenton[2], and Amy B. Pedersen[1]

[1]Department of Biological Sciences, University of Edinburgh, West Mains Rd, Edinburgh EH9 3JT, UK; [2]Institute of Integrative Biology, University of Liverpool, Crown Street, Liverpool L69 7ZB, UK

Using tools from community ecology to study patterns of disease has been useful in elucidating interactions with host resources, immune response, and between co-infecting parasites. These processes can lead to a non-random assembly of parasite communities, where communities of low species richness are often found to be a subset of high richness communities, a phenomenon known as nestedness. Nestedness occurs when certain species are found in both species-rich and species-poor communities, and suggests a particular order of immigration (infection, in the case of parasites) or dominance. However, although most individuals in wild populations are infected with multiple parasites, we have little knowledge about how within-host parasite communities are structured. We tested the nestedness of within-host communities of 7 taxonomically diverse parasite species infecting wild wood mice (*Apodemus sylvaticus*) in Northern England over three-years. *Bartonella* spp were the most prevalent parasites, being found in communities across all richness levels, followed by less prevalent parasites such as *Eimeria* spp, and *Heligmosomoides polygyrus*. We found evidence of significant nestedness in these within-host parasite communities, meaning that communities were not simply random assemblages of all possible parasites. These results inform predictions about the order in which infections occur, which are currently being tested using a long-term, longitudinal dataset of infections in these wild mouse populations. Our findings illustrate how parasite community patterns at a population level can be driven by individual exposure to parasites, and suggest that within-host parasite interactions may be important for both parasite dynamics and the host's immune response to co-infections.

12:15 PM - 12:30 PM (15 mins)

Effective networking across metabolic, immune, and gut microbial systems may be crucial to fight *Leishmania major* infection *

Sabrina Lamour, Kirill A. Veselkov[1], Joram M. Posma[1], Emilie Giraud[2], Matthew E. Rogers[3], Simon Croft[2], Julian Marchesi[4, 5], Elaine Holmes[1], Karin Seifert[2], and Jasmina Saric[1]

[1]Department of Surgery and Cancer, Imperial College London, SW7 2AZ, UK. [2]Department of Immunology and Infection, London School of Hygiene & Tropical Medicine (LSHTM), London, WC1E 7HT, UK. [3]Department of Disease Control, LSHTM, London, WC1E 7HT, UK. [4]Cardiff School of Biosciences, Division of Microbiology, Cardiff University, Cardiff, CF10 3AT, UK. [5]Centre for Digestive and Gut Health, Imperial College London, SW7 2AZ, UK.

The leishmaniases are diseases caused by protozoan parasites that are difficult to manage due to suboptimal diagnosis, variable clinical manifestations, and limited treatments. To understand the determinants of disease progression, we have performed a comprehensive characterisation and integration of multiple host parameters towards parasitic infection. We have, therefore, characterised two opposing mouse models of cutaneous leishmaniasis that are well-known for their distinctive disease phenotypes, namely self-healing C57BL/6 mice and non-healing BALB/c strain. We uniquely show that the two models display differential host responses to *Leishmania major* on a metabolic level, and also demonstrate clear differences in their inflammatory status and gut microbiota. Furthermore, we present a novel approach to integrate metabolic, immune, parasitic and microbial measures using correlation network analyses, a method that can be applied to other experimental and/or clinical data, in order to assess interactions and determinants of differential disease outcomes on a systemic scale. Our results revealed marked differences in interaction between host responses in the two models already a few days after infection. This rapid distinction implies that trans-molecular communication during the innate immune response is essential in mounting a successful systemic response towards *L. major* infection, a phenomena that may also prove to be important across other infectious diseases. Finally, our work also identifies a range of both parasitic- and host-derived metabolites that are linked with either successful or poor prognosis, providing useful biomarkers for the development of novel diagnostic tools for the complex diseases of *Leishmaniasis*.

Session F1 (RCJ) - Genomics of veterinary parasites

Chair: Matt Berriman - Wellcome Trust Sanger Institute

11:00 AM - 11:30 AM (30 mins)

Genomic and genetic approaches to investigate the molecular basis of anthelmintic resistance: *Haemonchus contortus* as a model system

John Gilleard
Faculty of Veterinary Medicine, University of Calgary.

Anthelmintic resistance is a major problem for the control livestock parasites and a potential threat to the sustainability of community-wide treatment programs being used to control human parasites in the developing world. Anthelmintic resistance is essentially a complex quantitative trait in which multiple mutations contribute to the resistance phenotype in an additive manner. Consequently, a combination of forward genetic and genomic approaches are needed to identify the causal mutations and quantify their contribution(s) to the resistance phenotype. Therefore, there is a need to develop genetic and genomic approaches for key parasite species identified as relevant models. *Haemonchus contortus*, a gastro-intestinal parasite of sheep, has shown a remarkable propensity to develop resistance to all the drugs used in its control. Partly because of this, and partly because of its experimental amenability, research on this parasite has contributed more than any other to our understanding of anthelmintic resistance. *H. contortus* offers a variety of advantages as an experimental system including the ability to undertake genetic crosses; a prerequisite for genetic mapping. In this presentation, I will discuss the current progress on developing *H. contortus* as a model system in which to undertake genetic and genomic approaches to study anthelmintic resistance

11:30 AM - 11:45 AM (15 mins)

Molecular epidemiology of *Leishmania donovani* in the Indian sub-continent: whole genome sequencing reveals bottlenecks, clonal outbreaks, migration and recombination

Jean-Claude Dujardin [1], Imamura H[1], Downing T[2], Mannaert A[1], Rijal S[3], Sundar S[4], Rai K[3], Vanaerschot M[1], Bhattarai N[3], Schonian G[5], Berriman M[6], Cotton J[6].
[1.] Institute of Tropical Medicine, Antwerp, Belgium [2.] NUI, Galway, Ireland [3.] BPKIHS, Dharan, Nepal [4.] BHU, Varanasi, India [5.] Charité U, Berlin, Germany [6.] WTSI, Hinxton, UK

Molecular epidemiology of visceral leishmaniasis (VL) in the Indian sub-continent (ISC) was long been hindered by the lack of resolution of molecular markers. Next generation sequencing is now providing the ultimate solution to this problem. We sequenced the whole genome of 203 *L. donovani* clinical isolates collected in the last decade in the ISC. We identified two completely distinct (45,743 SNPs) lineages: (i) The 'Yeti-12' is homogeneous (5628 SNPs), and tends to originate from an atypical, highland VL focus in Nepal; (ii) The 'Core-191' is endemic in the lowlands and likely emerged around 1850 (median estimation by BEAST analysis), is relatively homogeneous (2,418 SNPs), but was split in 9 clades, suggesting a recent 'evolutionary Big Bang' in the ISC, after the massive DDT campaign of the 1960s. Within several of these clades, a recent radiation was estimated to have occurred 50 years ago (95% CI, 38-67 yrs), probably during the first VL epidemics after the DDT campaign. Geo-localisation of the 9 sub-populations revealed local outbreaks and migratory patterns. Patterns of hybridization have also been detected. Our Indian sample set was collected entirely during the last VL epidemics and showed clonal expansion of limited genotypes during the rise of the epidemics and more genotypes during the fall. Simple PCR assays are now available to track the main genotypes of the ISC. Approaching the deadline of the VL elimination programme, it is critical to follow up the spatial and temporal distribution of these populations to better understand their impact on VL epidemiology.

11:45 AM - 12:00 PM (15 mins)

VSG-Seq: A quantitative method for analyzing *Trypanosoma brucei* Variant Surface Glycoprotein expression *in vivo* *

Monica Mugnier, George Cross, Nina Papavasiliou
The Rockefeller University, New York, New York, USA

Using a repertoire of over 2000 different variant surface glycoprotein (VSG) genes within its genome, *Trypanosoma brucei* changes its dense VSG surface coat to avoid detection by the immune system of its mammalian host. The dynamics of antigenic variation in *T. brucei* during an infection, however, are poorly understood. Unanswered questions include: How many variants appear over the course of an infection? Can the immune system of the host affect antigenic

variation *in vivo*? Is there a pattern to VSG expression over time? Although some of these questions have been broached using Sanger sequencing of VSG cDNA, technical limitations have prevented a high-resolution, quantitative study of VSG expression during *T. brucei* infection. Here we present the first method for quantitatively examining the diversity of expressed VSGs in a population of trypanosomes, isolated either from culture or from blood. This next-generation sequencing approach requires very little input material and is quite sensitive, detecting VSGs expressed on less than 0.1% of a population of trypanosomes. Using samples isolated from mouse infections, expressed VSG sequences can be assembled accurately *de novo*, demonstrating that this approach can be used for the high-resolution study of VSG expression in any strain of *T. brucei*, whether in the lab or in the field.

12:00 PM - 12:15 PM (15 mins)

Harnessing related species and samples data to create and optimise draft genome sequences for *Leishmania* species *

Simone Coughlan[1], Gabriele Schonian[2], Matthew Berriman[3], Tim Downing[1].

[1.]School of Mathematics, Applied Mathematics and Statistics, National University of Ireland, Galway, Ireland [2.]Charite University, Berlin, Germany; [3.]Wellcome Trust Sanger Institute, Hinxton, UK

Effective molecular tools for monitoring the emergence of novel pathogens in domestic and peridomestic reservoir hosts are urgently required. Taxonomically classifying unknown samples of ambiguous origin and identifying optimal protocols for their genome assembly using short-read data is required for comparison with known species. DNA was sampled from two Colombian dogs with leishmaniasis and from an Ethiopian rat harboring a new species called *L. arvicanthis*. These samples were sequenced to produce paired-end short read Illumina libraries. A genome assembly pipeline that iteratively optimised and transformed the short read data into high quality draft genomes was developed, including the current *L. braziliensis* genome data as a reference control. Phylogenetic markers were extracted from the genomes and compared with markers from a panel of known *Leishmania* species, identifying two of them as members of the *L. braziliensis* complex: *L. naiffi* and *L. shawi*. The assembly pipeline implemented here involves stringent QC filtering, de novo assembly, iterative gap-filling and base correction steps before identifying and removing potential mis-assemblies. Then, using reference genome data (*L. braziliensis* and *L. tarentolae* here), it aligns, orders and orients scaffolds into pseudo-chromosomes and transfers the reference annotation onto the new draft genome. Short read coverage and allelic diversity determined variation across four levels in the draft genomes: ploidy, whole chromosome copy number, structural changes and SNPs. Although *L. naiffi* and *L. shawi* were triploid, *L. arvicanthis* was diploid, and aneuploidy was also observed in all three species, highlighting the universality of multi-levelled genome plasticity in differing environments.

12:15 PM - 12:30 PM (15 mins)

Application of Reverse Line Blotting to the study of tick-borne haemoparasites - A West African Experience *

Vincenzo Lorusso[1,2], Michiel Wijnveld[3], Ayodele Majekodunmi[1], Augustine Igweh[4], Frans Jongejan[3], Susan C Welburn[1], Kim Picozzi[1]

[1.]Division of Pathway Medicine, Edinburgh University Medical School, The Chancellor's Building, 49 Little France Crescent, Edinburgh EH16 4SB, United Kingdom [2.]VETOQUINOL Laboratories, 37 Rue de la Victoire, 75009 Paris, France [3.]Utrecht Centre for Tick-borne Diseases (UCTD), Faculty of Veterinary Medicine, Utrecht Universiteit, 1 Yalelaan 3584CL, Utrecht, The Netherlands [4.]Nigerian Institute of Trypanosomiasis Research (NITR), VOM, Jos, Plateau State, Nigeria

Tick-borne haemoparasites (TBHs) threaten livestock health, welfare, and production in Africa. Based on the hybridization of PCR products to probes linked to a membrane, Reverse Line Blotting (RLB) can enable the simultaneous detection of several TBHs. This presentation describes the implementation and the application of an RLB method optimized for the diagnosis of TBHs of veterinary and zoonotic importance in a West African setting, where no molecular survey had ever been conducted before. Following DNA extraction, three simultaneous PCRs targeting *Ehrlichia/Anaplasma*, *Theileria/Babesia*, and *Rickettsia* spp. were carried out on whole blood samples collected from indigenous (*Bos indicus*) cattle from Plateau State, central Nigeria, and from whole blood samples collected from camels (*Camelus dromedarius*) in Sokoto, north-western Nigeria. PCR products were then screened against a panel of five genus-specific and 12 species-specific probes. Samples positive only for genus-specific probes were subjected to purification and sequence analysis. RLB proved to be a versatile and sensitive technique, detecting also sub-clinical infections otherwise overlooked. More than 70 different species combinations were found infecting cattle. New haemoparasite-vertebrate host relationships were disclosed. Amongst others, cattle harbored *Anaplasma platys* and *Rickettsia massiliae*, providing the first report of detection of this zoonotic rickettsia in a vertebrate host. Camels were found infected for the first time with *A. platys* and *Theileria ovis*. According to the results obtained, a further RLB was set up, comprising five genus-specific and 25 species-specific probes, suitable for epidemiological and case-control studies to be carried out across the whole of Africa.

Session A2 (WRL) - Kinetoplastida - Gene expression

Chair: Miguel Navarro, IPBLN - Spanish National Research Council

2:00 PM - 2:30 PM (30 mins)
Gene expression in trypanosomes - the control freak's guide
Isabel Roditi
Institute of Cell Biology, University of Bern, Bern, Switzerland.

Post-transcriptional regulation, in particular RNA stability, has long been regarded as the central mechanism for regulating gene expression by trypanosomes. It has been known for some time that the parasites employ a much larger set of control mechanisms to regulate expression of the major surface proteins, which are transcribed by RNA polymerase I. These include regulation of transcription initiation, elongation, RNA maturation and translation. Recent genome-wide analyses indicate that multiple layers of regulation are the rule, not the exception, and that chromatin structure and co-transcriptional control mechanisms also contribute to determining expression levels of messenger RNAs from the polycistronic clusters transcribed by RNA polymerase II. At the next level, cohorts of transcripts, whose genes are dispersed throughout the genome, can form regulons whose RNA stability or translation are coordinately regulated. Relatively few regulatory proteins have been identified to date. Genome-wide RNAi screens and comparative RNA-Seq have recently led to the identification of novel regulators of stage-specific expression. These include conserved housekeeping proteins that seem to have acquired additional functions in trypanosomes.

2:30 PM - 2:45 PM (15 mins)
Chromatin readers regulate monoallelic expression and switching in *Trypanosoma brucei*.
Danae Schulz, Danae Schulz, Erik Debler, Monica Mugnier, George Cross, Nina Papavasiliou
Rockefeller University 1230 York Ave. New York, NY 10065

The protozoan parasite *Trypanosoma brucei* is the causative agent of Human African Trypanosomiasis. The *T. brucei* surface is covered by a coat made of Variant Surface Glycoprotein (VSG). There are ~2000 VSG genes in the genome, but only one is expressed at a time. *T. brucei* evades the host immune system through antigenic variation, periodically switching the expressed VSG. This requires tight control of both monoallelic VSG expression and switching frequency. VSGs are expressed from one of ~15 telomeric expression sites (ESs) while the rest are transcriptionally silenced. The molecular mechanisms that control monoallelic expression and switching are largely unknown, but many studies demonstrate that chromatin factors play a role. I tested whether bromodomain-containing proteins, a subclass of chromatin proteins that 'read' the histone code, maintain monoallelic expression of VSG genes. Treating trypanosomes with the bromodomain inhibitor iBET151A results in cell cycle arrest, transcription at silent ESs, expression of multiple VSGs simultaneously, and a 100-fold increase in switching frequency. The bromodomain protein Bdf2 binds to iBET151A, and knockout of Bdf2 phenocopies treatment with the inhibitor. RNA-seq analysis indicates that Bdf2 primarily affects transcripts of genes involved in immune evasion and pathogenesis, while ChIP demonstrates Bdf2 localization to silent and active ESs. Current efforts are focused on a Bdf2-iBET151A crystal structure with the aim of designing a more trypanosome-specific inhibitor for therapeutic intervention. Our studies indicate that bromodomain proteins are important for maintaining monoallelic expression and regulating switching frequency, and provide insight into the mechanism of immune evasion for this deadly parasite.

2:45 PM - 3:00 PM (15 mins
Whole transcriptome analysis of *Leishmania major* virulence factors
Vyacheslav Yurchenko[1,3], Aygul Ishemgulova[1], Jan Votýpka[2,3], Pavel Flegontov[1,3], Petr Volf[2],
[1.] Life Science Research Centre, Faculty of Sciences, University of Ostrava, 710 00, Ostrava, Czech Republic; [2.] Department of Parasitology, Faculty of Science, Charles University, 128 44 Prague, Czech Republic; [3.] Biology Centre, Institute of Parasitology, Czech Academy of Sciences, 370 05 České Budějovice (Budweis), Czech Republic

Over the years many genes have been implicated in *Leishmania* pathogenicity. In this work we compared the expression profiles of virulent and avirulent strains of *L. major* using next-generation sequencing. Long-term cultivation of the *L. major* virulent strain LV561 (LRC-L137; MHOM/IL/1967/Jericho-II) yielded an avirulent line LV561/AV with attenuated infectivity for mice and sand fly vectors. Such reduced virulence was primarily explained by the differences in LPG and

GP63 expression on the cell surface, but several other molecules (e.g. NADPH-diaphorase and peroxidase) were also considered to be involved. Whole transcriptome analysis of polyA RNA revealed that 230 and 357 transcripts are upregulated in LV561/V (virulent) and LV561/AV (avirulent) strains, respectively ($p \leq 0.05$). Eleven out of the top 50 genes with elevated expression in LV561/V were localized to chromosome 19. This disproportion is even more staggering in the case of the LV561/AV, where 29 out of the top 50 upregulated transcripts originated from chromosome 35. Among differentially expressed genes we identified those involved in cell attachment, membrane dynamics, protection from the oxidative stress, glucose and folate transport, proteases and others. Interestingly, a significant proportion of genes with elevated expression in LV561/AV may be involved in chromatin remodeling and RNA processing suggesting that attenuation of pathogenicity is directly linked to gene expression.

3:00 PM - 3:15 PM (15 mins

Allelic exclusion by VEX1 controls antigenic variation in trypanosomes

Lucy Glover and David Horn
College of Life Sciences, University of Dundee, Dundee

Antigenic variation in African trypanosomes requires monoallelic transcription of a single telomeric Variant Surface Glycoprotein (VSG) gene and the concomitant reversible silencing of a repository of VSGs at other telomeres. The active VSG colocalises with a concentration of RNA polymerase-I at an extranucleolar focus known as the expression-site body or ESB. Although a factor that displays specificity for the active allele has been suspected as a regulator of VSG expression, no such factor has been identified. To search for a protein in this class, we conducted a genome-scale RNA interference library screen for loss of telomeric gene silencing in bloodstream-form trypanosomes. This screen identified a novel gene required for VSG gene regulation. Knockdown targeting this gene resulted in loss of telomeric VSG silencing, producing cells coated with multiple VSGs. This protein, named VSG EXclusion 1 or VEX1, was found concentrated into a focus adjacent to the ESB. Notably, while the ESB was lost, the VEX1 focus persisted in insect-stage cells, which do not transcribe VSGs. We conclude that VEX1 is a monoallelic regulator that restricts VSG transcription to a single telomere, underpinning antigenic variation and host immune evasion in the African trypanosome.

3:15 PM - 3:30 PM (15 mins)

A nucleolar protein of *Trypanosoma brucei* is involved in both surface protein and ribosomal RNA expression *

Salome Aeschlimann and Bernd Schimanski
Institute of Cell Biology, University Bern, Baltzerstrasse 4 3012 Bern Switzerland

Nuclear mRNA export is a key step in the cascade of interconnected processes of gene expression. Main players involved in the translocation of mature mRNAs are well described in higher eukaryotes. However, very little is known about the export machinery in *Trypanosoma brucei*. Only a few export factors are characterized - among them the ortholog of Sub2, a member of the transcription-export (TREX) complex in higher eukaryotes. Immunoprecipitation of TbSub2 of procyclic forms led to the identification of Tb927.10.9130. This protein is annotated as a putative pre-mRNA splicing factor and we could show that it localizes to the nucleolus. In all eukaryotes the nucleolus is the site of rRNA transcription by RNA polymerase I. In addition to that, *T. brucei* uses RNA pol I for the transcription of the major surface protein genes in both life cycle stages. Interestingly, partial downregulation of Tb927.10.9130 by RNAi had no influence on growth but led to a decrease of procyclin indicating a role in the expression of protein coding genes transcribed by RNA pol I. Moreover, RNAi in bloodstream forms -although inefficient - led to lower VSG levels and had no influcence on cell viability. In contrast, complete loss of the protein after conditional knockout resulted in growth inhibition due to decreased levels of 18S rRNA indicating a second function for Tb927.10.9130. An effect on bulk mRNA distribution was not observed. Currently, we are analyzing interacting proteins to gain more insight into the dual roles of Tb927.10.9130, and especially in surface coat expression.

Session B2 (WRR) - Apicomplexa - Mosquitoes and sexual stages

Chair: Jacob Baum, Walter & Eliza Hall Institute of Medical Research

2:00 PM - 2:30 PM (30 mins)
No title
Martin Donnelly
Liverpool School of Tropical Medicine

Abstract not received

2:30 PM - 3:05 PM (20 mins)
Role of the apiAP2 proteins in the life cycle of the malaria parasite.
Katarzyna Modrzynska, Claudia Pfander, Lia Chappell, Matt Berriman, Oliver Billker
Wellcome Trust Sanger Institute

The majority of the transcription machinery is highly conserved between *Plasmodium* and other Eukaryota. However, the scarcity of predicted transcription factors in the *Plasmodium* genome suggests that the gene specific control mechanisms are quite divergent between the parasite and its host. The plant-like ApiAP2 proteins constitute the largest family of putative transcription factors found in the *Plasmodium* genome and have been suggested to be the key regulators of parasite development. Their exact function and specificity however remain to be elucidated. We have therefore conducted a systematic gene knock-out screen, targeting all 26 member of the apiAP2 family in the rodent parasite *Plasmodium berghei*. Twelve clonal lines could be generated and phenotyped throughout the full life cycle. We observed an array of previously undescribed phenotypes including defects in gametocytogenesis (2 lines), in ookinete formation (4 lines), and in oocyst development and maturation (3 lines). We have also used directional RNA-seq to study the changes in transcriptome of all 12 mutants at schizont gametocyte and ookinete stages. Similar defects, such as loss of gametocytes, were often associated with different transcription patterns that provided further clues about the nature of the observed phenotypes i.e. defect of gametocyte commitment in one mutant vs. overexpression of normally silent genes interfering with the gametocyte development in another. This global analysis of function of all targetable members of the apiAP2 family contributed to our understanding of the molecular mechanism of transcription regulation in *Plasmodium* and its role in malaria life cycle.

3:05 PM - 3:25 PM (20 mins)
High throughput reverse genetics screening in *Plasmodium berghei* using signature-tagged mutagenesis unravels genetic interactions *
Ana Rita Gomes, Ellen S Bushell, Mathieu Brochet, Frank Schwach, Gareth Girling, Michael Quail, Burcu Bronner-Anar, Mandy Sanders, Christian Doerig[1], Julian Rayner, Oliver Billker
Wellcome Trust Sanger Institute, Cambridge, UK and [1]Monash University, Melbourne, Australia

Signature-tagged mutagenesis (STM) has been used extensively in bacterial pathogens to identify virulence genes by parallel phenotyping of pools of barcoded mutants. Here we demonstrate that the increased recombination frequency of PlasmoGEM vectors, combined with a strongly reduced incidence of episomes and the presence of a gene-specific tag or barcode, now make it possible to reproducibly generate pools of over 50 targeted knockout (KO) mutants in a single mouse. The barcodes carried by each mutant can be quantified by next generation sequencing, which allowed us to calculate the relative growth rate for each mutant within the pool and how it changes during the infection. To identify potential interaction pairs within the *P. berghei* kinome we performed STM experiments in seven different mutant lines. The data generated revealed multiple growth phenotypes that were recurrent in all backgrounds and associated with different stages of the infection. Additionally, a severe growth defect was detected for a mutant lacking the CDPK4 gene on a line expressing the resistant pkgT619Q allele, thus suggesting the existence of an important genetic interaction between CDPK4 and PKG that has also been validated independently. We demonstrate how, with our *Plasmodium*-specific approach to STM, the growing PlasmoGEM resource can be used in systematic genome-wide screens to identify parasite genes that are redundant for asexual growth as well as to perform genetic interaction screens. This kind of high throughput genetic approach has no precedents in the malaria field and has revealed a link between PKG and calcium metabolism.

Session C2 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment II

Chair: Jeremy Sternberg, University of Aberdeen

2:00 PM - 2:30 PM (30 mins)
Chagas' disease: progress and challenges.
Michael Miles
London School of Hygiene and Tropical Medicine

Abstract not received.

2:30 PM - 2:45 PM (15 mins)
Global Distribution Maps of the Leishmaniases *
David Pigott[1], Samir Bhatt[1] Nick Golding[1] Kirsten A Duda[1] Katherine E Battle[1] Oliver J Brady[1] Jane P Messina[1] Yves Balard[2, 3] Patrick Bastien[2, 3] Francine Pratlong[2, 3] Susan Aman[4] John S Brownstein[4, 5] Clark Freifeld[4, 6] Peter W Gething[1] Dylan B George[7] Monica F Myers[1] Richard Reithinger[8] Simon I Hay[1, 7]
[1]Spatial Ecology and Epidemiology Group, Tinbergen Building, Department of Zoology, University of Oxford, South Parks Road, Oxford, United Kingdom; [2]University Montpellier 1 (UFR Médecine) & CNRS 5290/IRD 224 (UMR "MiVEGEC"), Laboratoire de Parasitologie-Mycologie, Montpellier, France; [3]CHRU de Montpellier, Centre National de Référence des Leishmanioses, Department of Parasitology – Mycology, 39, Avenue Charles Flahault, 34295 Montpellier Cedex 5, France; [4]Children's Hospital Informatics Program, Boston Children's Hospital, Boston, MA, United States of America; [5]Department of Pediatrics, Harvard Medical School, Boston, MA, United States of America; [6]Department of Biomedical Engineering, Boston University, Boston, MA, United States of America; [7]Fogarty International Center, National Institutes of Health, Bethesda, MD 20892, United States of America; [8]RTI International, Washington D.C., United States of America.

The Leishmaniases are a collection of complex infections caused by *Leishmania* spp., ranging from localised cutaneous lesions to forms with visceral complications. Annual incidence is estimated to be around 2 million new cases. The interplay between humans, the Phlebotomine sandfly vectors and reservoir hosts complicates epidemiological understanding as well as control efforts; research has therefore tended to concentrate on solving clinical and epidemiological aspects of the disease at small spatial scales. As a result, no attempt has been made to provide a global evidence-based risk map of these diseases. For each sub-national province, an assessment of cutaneous and visceral leishmaniasis was performed incorporating data from the WHO Expert Committee and GIDEON as well as peer-reviewed disease occurrences and reported annual caseloads. These data were used to quantitatively assess certainty of the diseases' presence or absence on a continuous scale. A global database of 12,500 geo-positioned data points was then collected from peer-reviewed literature and other sources. Using a Boosted Regression Trees modelling approach, separate continuous global environmental risk maps for Cutaneous and Visceral Leishmaniasis were produced. Climatic and environmental variables were identified as important in defining this distribution. These risk maps were also converted into sub-national estimates of incidence. It is hoped that such maps will help inform not only future epidemiological studies but also public health policy directed towards these diseases, allowing improved targeting of specific control efforts with humans, vectors and reservoirs, as well as identify suitable areas for surveillance both active and passive.

2:45 PM - 3:00 PM (15 mins)
Trypanosomiasis in domestic livestock in the Luangwa valley in Zambia. *
Kathrin Schaten, Schaten K., Simuunza M., Machila N., MacLeod E., Shaw A., Thrusfield M., Welburn S.
University of Edinburgh, Department of Pathway Medicine, Royal Infirmary of Edinburgh, Chancellor's Building, Edinburgh

The Luangwa valley in Zambia is historically endemic for African trypanosomiasis, but migration and land use change that have occurred over the last 20 years, may have altered the prevalence of both sleeping sickness and nagana in this area. The proximity to the Luangwa National Park means that this area still has a relatively dense population of wildlife that

may act as a reservoir for trypanosomiasis in animals and humans. In 2012, a census was conducted to identify the number of people and animals living in the study area. In total, 3,700 households were identified. Out of these, 140 households were selected for the animal survey done between June and August 2013. In each selected household, every cow, goat, sheep, pig and dog were sampled, provided consent was given. Samples were stored on FTA cards for analysis at the University of Edinburgh Molecular analysis shows that around 25% of cattle are positive for trypanosomes. The other tested livestock species are also affected, but much less than cattle while very few dogs tested positive so far. This study does not only try to assess the prevalence of trypanosomiasis in the valley but also the possible impact of keeping several livestock species together and discuss the role of a sentinel species for the risk of human African trypanosomiasis.

3:00 PM - 3:15 PM (15 mins)

Mapping the incidence and detection probability of human African trypanosomiasis

Nick Golding

Spatial Ecology and Epidemiology Group, Department of Zoology, University of Oxford, South Parks Road, Oxford OX1 3PS, UK

Effective planning for the control and eventual elimination of human African Trypanosomiasis (HAT) will require accurate and reliable spatial information on the distribution of the disease. The Atlas of HAT, recently established by WHO and FAO, aims to map the locations of all known HAT cases. Whilst these data provide an unparalleled resource for spatial risk assessment, the strong spatial bias in case reporting necessitates the development of predictive models to produce continuous maps of disease risk. Unfortunately the complexity of the dataset (which contains case reports from both active and passive detection) and the highly focal nature of the disease limit the applicability of existing spatial models. To overcome these issues, a collaboration between the Spatial Ecology and Epidemiology Group, WHO and FAO is developing a novel Bayesian modelling framework to simultaneously map the incidence of HAT cases (both *gambiense* and *rhodesiense* forms) and the probability of detection of cases. We describe the suite of sociological and environmental correlates developed to map these two processes, outline the joint statistical model developed and present preliminary results. The resulting maps will provide accurate subnational estimates of the incidence of HAT to guide the implementation of interventions, highlight areas where case detection could be improved and identify regions where previously undetected transmission foci may be present.

3:15 PM - 3:30 PM (15 mins)

Human migration drives the dispersal of epizootic Chagas disease: the case of highland Bolivia *

Louisa Messenger[1], Lineth Garcia[2], Michael A. Miles[1] and Martin S. Llewellyn[1]

[1]London School of Hygiene and Tropical Medicine, London, United Kingdom; [2]Universidad Mayor de San Simón, Cochabamba, Bolivia

Trypanosoma cruzi, the aetiological agent of Chagas disease, is an ancient and widespread zoonosis distributed throughout the Americas. TcI is the most abundant and expansive *T. cruzi* genetic lineage; it is the principal cause of human chagasic cardiomyopathy in Colombia and Venezuela and is ubiquitous among sylvatic transmission cycles. Multiple molecular markers consistently identify high levels of genetic variation within sylvatic TcI populations, and divergent, but genetically homogeneous, strains isolated from human infections. However, current understanding of the genetic determinants that drive natural *T. cruzi* diversification is incomplete. We performed high resolution nuclear and mitochondrial genotyping of contemporaneous sylvatic TcI (n=199 biological clones), isolated from a range of triatomine and mammalian hosts across Bolivia. We detected two distinct sylvatic transmission cycles in adjacent highland and lowland areas. Highland Bolivian strains were characterized by reduced genetic diversity and heterozygosity (Ar= 1.92-2.22, FIS=-0.241-0.026) compared to lowland areas (Ar = 3.40-3.93, FIS=0.176). We observed equivalent levels of subdivision among highland areas spanning >465 km (FST = 0.084) and between lowland populations across 155 km (FST = 0.084). Measurements of isolation by distance detected greater parasite dispersal among geographically disparate highland areas (RXY= 0.053, p=0.142) than between proximate lowland foci (RXY= 0.209, p<0.001). The most parsimonious explanation for our results is a founder event in highland Bolivia with long-range anthropogenic dispersal of parasites across an ecological cline. We discuss the role of humans as an abundant, but often neglected, vector of *T. cruzi* and consider their implications for the epidemiological risk of emergent epizootic Chagas disease.

Session D2 (RCA) - Helminths - Molecular epidemiology and transmission

Chair: David Rollinson, The Natural History Museum

2:00 PM - 2:30 PM (30 mins)

Achieving Transmission Control of Schistosomiasis: Is There a Role for Biological Enemies of Snails or Schistosomes?

Eric Loker, Martina Laidemitt, Martin W. Mutuku1, Sarah Buddenborg, Lijun Li, Si Ming Zhang, Gerald M. Mkoji.1
Department of Biology, Center for Evolutionary and Theoretical Immunology, University of New Mexico, Albuquerque, New Mexico, USA 87131 1. Center for Biotechnology Research and Development, Kenya Medical Research Institute, Mbagathi Road, P.O. Box 54840-00200, City Square, Nairobi, Kenya

The call for the global elimination of schistosomiasis as a public health problem by 2025 carries with it many significant challenges, including the need to augment praziquantel treatment such that transmission control can be achieved, and prevalence can be more sustainably reduced. Control of schistosome-transmitting snails, or of schistosome sporocysts within them, offers one way to achieve this goal. There is surely a place for use of molluscicidal chemicals or for environmental control (drainage, vegetation removal, etc.) in some contexts. But what about the use of natural enemies, whether they be pathogens, competitors, or predators of either snails or, preferably, schistosome sporocysts? In general, there has been little recent effort to find such enemies, explore their properties, or to contemplate whether there are practical and safe ways to use them. Additionally, to what extent do such enemies quietly already play a role in limiting the number of infections occurring in snails? Other general factors to consider here are 1) to what extent will the ongoing introduction of exotic species into schistosome-transmitting habitats – whether we approve or not – influence transmission in some places; and 2) will degradation of aquatic habitats to the point that schistosome-transmitting snail populations can not survive provide a rather sad form of transmission control? Recent approaches for identifying and investigating natural enemies as potential control agents will be highlighted, including some thoughts for how they might be deployed. Among the organisms we are examining are amphistome flukes and Daubaylia nematodes

2:30 PM - 2:45 PM (15 mins)

Population genetic studies of *Schistosoma haematobium* using novel multiplex microsatellites: inter and intra species molecular epidemiology

Bonnie Webster[1, 2], Muriel Rabone[2], Fiona Allan[2], Aidan Emery[2], Joanne Webster[1], Amadou Garba[3], David Rollinson[1]
[1.]Department of Infectious Disease Epidemiology, Faculty of Medicine, Imperial College (St Mary's Campus), Norfolk Place, London W2 1PG, UK; [2.]Department of Life Sciences, Parasites and Vectors Division, Natural History Museum, Cromwell Road, London, SW7 5BD, UK; [3.]Reseau International Schistosomoses, Environment, Amenagements et Lutte (RISEAL) Niamey, Niger

The Schistosomiasis Consortium for Operational Research and Evaluation (SCORE) is a multi-centre collaborative project to answer strategic questions about schistosomiasis control and elimination. Advances in DNA technology, together with the recent development of high-throughput and cost effective methods, to capture schistosome larval stages from endemic areas and to obtain DNA from samples stored on Whatman FTA® cards have revolutionised the way we can analyse schistosome populations sampled directly from their natural hosts. One of the SCORE research aims is to evaluate the effect of different Mass Drug Administration (MDA) regimes on the population genetics of *Schistosoma haematobium*. For this over a million random *S. haematobium* DNA fragments were sequenced to create a new suite (13,000) of unique microsatellite population genetic markers from which 15 were chosen and fully optimized. These 15 together with 3 pre-existing loci have now been developed into two robust novel multiplex PCR's to investigate the genetic variation and population structure of *S. haematobium* larval stages collected within the SCORE project. Here we present microsatellite data from individual *S. haematobium* miracidia harvested from infected human urine samples collected from mainland Africa and Zanzibar, and the data are discussed in relation to: 1. The distinct mitochondrial genetic separation previously identified between *S. haematobium* from the Indian Ocean Islands and from the African mainland, and 2. The complications that inter-species interactions could have on the interpretation of microsatellite data from areas where closely related *S. haematobium*-group species are transmitted sympatrically.

2:45 PM - 3:00 PM (15 mins)

Distribution of Tetraspanin-23 alleles in hybrid schistosomes.

Tine Huyse[1], Nele Boon[2,3], Frederik Van den Broeck[2,3], Filip Volckaert[2], Katja Polman[3]

[1.]Section Invertebrates, Royal Museum for Central Africa, Leuvensesteenweg 13, 3080 Tervuren, Belgium. [2.]Laboratory of Biodiversity and Evolutionary Genomics, Biology, University of Leuven, Belgium [3.]Unit of Medical Helminthology, Institute of Tropical Medicine, Antwerp, Belgium

To date, only neutrally evolving genes have been studied in the context of hybridization and introgression in schistosomes. Here we chose to study tetraspanin-23 (TSP-23) as this membrane-bound protein is thought to interact with the immune system, and has been shown to be under positive selection in schistosomes. We studied the genetic diversity of the tetraspanin-23 gene in various *S. haematobium* x *S. bovis* and *S. haematobium* x *S. mansoni* crosses collected in northern Senegal. These results were compared with the 'neutral' ITS rDNA and partial cox1 sequences to test for asymmetrical gene flow. The observed patterns of allele distribution and introgression will be discussed.

3:00 PM - 3:15 PM (15 mins)

Look to the snails: indicators of schistosomiasis transmission within control programmes

Fiona Allan

Department of Life Sciences, The Natural History Museum, Cromwell Road, London, SW7 5BD

Schistosomiasis is a disease with focal transmission centred around water bodies containing susceptible intermediate snail hosts. Prevalence of schistosomiasis in the human population is being reduced by mass drug administration (MDA), increasing the need to monitor transmission to assess the impact of control programmes; particularly in areas where control progresses towards elimination. SCORE is a consortium programme undertaking operational research on schistosomiasis control. Snail monitoring is on-going around villages under different control regimes, for *Schistosoma mansoni* in Tanzania and for *S. haematobium* in Niger as part of SCORE "Gaining and Sustaining" studies. *Bulinus* spp. snails are collected on a monthly basis in Niger, and *Biomphalaria* spp. snails on a quarterly basis in Tanzania. Abundance and patent infection prevalence data have been recorded for nearly 3 years. Species of snails and schistosome cercariae are identified in the field by morphology and a subset confirmed by molecular barcoding at the Natural History Museum. In addition, as part of the Zanzibar Elimination of Schistosomiasis Transmission (ZEST) programme, 15 shehias (administrative wards) on both Unguja and Pemba islands are targets of snail monitoring and control (with niclosamide). Approximately 200 transmission sites have been surveyed and treated so far. Data on snail density and parasite prevalence will be linked with parasitological surveys of school age children and used to assess the effectiveness of control interventions.

3:15 PM - 3:30 PM (15 mins)

Pattern of soil-transmitted helminth re-infections among Orang Asli schoolchildren in Malaysia

Hesham Al-Mekhlafi, Al-Delaimy AK, Lim YAL, Mahmud R

Department of Parasitology, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur, Malaysia

Despite the continuous efforts to improve the quality of life of Orang Asli (aborigines) communities in rural Malaysia, the trend of soil-transmitted helminth (STH) infections remains largely unchanged since the 1920s, with alarming high prevalence rates and prominent morbidity. This longitudinal study aimed at investigating the pattern of STH re-infections (incidence and intensity) among Orang Asli schoolchildren in Pahang, Malaysia. At baseline, faecal samples were collected from 145 schoolchildren and examined for STH by using formalin-ether sedimentation, Kato Katz and Harada Mori techniques. All infected children were treated with a 3-day course of 400 mg/daily of albendazole tablets and then re-examined monthly for 6 months. Almost all children (99.3%) were found to be infected by at least one STH species. The prevalence of trichuriasis, ascariasis and hookworm infections were 97.2%, 50.3% and 31.0%, respectively. Almost three-quarters and half of the trichuriasis and ascariasis, respectively, were of moderate-to-heavy intensities while all hookworm infections were of light intensity. After complete deworming, the re-infection with *Trichuris, Ascaris* and hookworm occurred rapidly and reached 50% of the baseline prevalence by second, third and fifth month, respectively. By 6 months, 80.7%, 41.4% and 23.4% of the children were re-infected by *Trichuris, Ascaris* and hookworm, respectively. Similar results were reported for the intensity of these infections. In conclusion, STH infections are still highly prevalent among Malaysian Orang Asli schoolchildren and this supports an urgent need to start integrated and effective STH control measures in order to reduce these infections significantly in these communities.

Session E2 (RCU) - Ecology - Local adaptation and coevolution

Chair: Olivier Restif, University of Cambridge

2:00 PM - 2:30 PM (30 mins)
Environmental stochasticity affects epidemics and host-parasite coevolution
Oliver Kaltz
Université Montpellier
All environments vary, and this often in unpredictable ways. We still know little about how environmental stochasticity affects the spread of epidemics, and even less about the consequences for the (co)evolutionary interactions between hosts and parasites. Classic epidemiological comparative approaches can be complemented by experiments, using miniaturised host-parasite systems in replicated microcosm populations, under controlled environmental conditions. I will present several experiments with a protozoan and a bacterial model system, where we varied the intensity of population disturbance or the spatio-temporal synchrony of temperature fluctuations. Our results indicate that changes in the mean, but also in the variance, of environmental conditions can impact outbreak thresholds and shifts in epidemic peaks, but also the long-term coevolutionary dynamics and the evolution of virulence.

2:30 PM - 2:45 PM (15 mins)
Host colour change and scent affect survival of the nematode *Heterorhabditis bacteriophora* *
Rebecca Jones, Andy Fenton, Mike Speed
Institute of Integrative Biology, University of Liverpool

Entomopathogenic nematodes belonging to the family *Heterorhabditidae* are lethal obligatory parasites of insects that are found in soil. These nematodes have direct life cycles, typically infecting insect larvae via a third stage infective juvenile (IJ) stage, which carries a symbiotic bacteria, *Photorhabdus* sp. These bacteria kill the host within 48 hours, following which the nematodes undergo a number of generations within the host before emerging as infective juveniles after a couple of weeks. What is striking is that hosts infected with *Heterorhabditid* nematodes undergo a dramatic colour change, becoming a pink-red in colour and also exhibiting bioluminescence. Although there are a number of hypotheses, the reasons for these changes in appearance of the infected cadaver are unknown. My research is focused on the hypothesis that the host changes colour as a parasite-induced warning signal to deter predators from consuming the host. Such predation would result in death of the infecting nematodes, and so this warning signal would therefore enable survival and propagation of the nematode-bacterium complex. Additionally, there seems to be a warning odour aspect to this avoidance as infected individuals have an unpleasant smell. My talk will focus on some recent experiments I have undertaken which examine this hypothesis and the combined roles of visual and odour cues as predator deterrents within this system.

2:45 PM - 3:00 PM (15 mins)
Assessment and Management of emerging nematode pests of Northern Ireland grassland and cereals
Thomas Fleming
Molecular Biosciences: Parasitology, School of Biological Sciences, Queen's University Belfast, 97 Lisburn Road, Belfast BT9 7BL; Pest Molecular Biology Group, Newforge Lane, Agri-Food Biosciences Institute, Belfast, BT9 5PX.

Plant parasitic nematode (PPN) damage in pastures and cereals is often subtle and difficult to detect, however when combined with abiotic stresses, crop damage can be significant. There is currently little information on the PPN species present in Northern Ireland (NI) pasture and cereal fields, but given the reliance of NI agriculture on these crops and the expected future increases in average soil temperature, the impact of these parasites is of increasing concern. This research project will determine which PPNs are currently causing crop damage and identify those species likely to emerge as more serious pests in the future. Initial sampling data across a range of soil types has shown a diverse range of PPNs in agricultural land, including economically important species such as root knot nematodes (*Meloidogyne naasi* and *M. minor*) at unexpectedly high occurrence levels. The occurrence of these endoparasites appears to be increasing, highlighting them as the most likely threats to future crop production. Management of PPNs in pasture and cereal crops will be investigated using a range of approaches including plant resistance, biostimulants and fungal endophytes. Initial results on the use of biostimulants in enhancing growth in stressed cereals and grasses will be presented.

3:00 PM - 3:15 PM (15 mins)

Bayesian modelling of factors potentially influencing the spatial distribution of *Echinococcus multilocularis* in foxes

Franz J. Conraths[1], Schwarz, S[1], Sutor, A[1], Hoffmann, L[2], Tackmann, K[1], Schmid, V[3], Staubach, C[1]

[1.] Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health, Südufer 10, 17498 Greifswald-Insel Riems, Germany; [2.] Thuringian State Authority for Food Safety and Consumer Protection, Tennstedter Str. 8/9, 99947 Bad Langensalza, Germany; [3.] Department of Statistics, Ludwig-Maximilians-University, Ludwigstr. 33, 80539 München, Germany

Alveolar echinococcosis is considered the most dangerous autochthonous parasitic zoonoses in central Europe. The red fox (*Vulpes vulpes*) represents the main definitive host in Europe and various rodent species are involved in the parasitic cycle as intermediate hosts. Since soil condition, temperature, and desiccation can reduce the infectivity of oncospheres, microclimate and habitat may represent factors that influence the spatial heterogeneity of the infection of foxes with *E. multilocularis*. To investigate the influence of environmental factors on the spatial epidemiology of *E. multilocularis*, 38,446 foxes were sampled in two Federal States of Germany and the results of the parasitological examination linked to a geographic information system. The landscape composition per spatial unit was derived from a high-resolution land-survey vector database and supplemented by a digital elevation model. Data were analyzed using a hierarchical Bayesian model in conjunction with Markov Chain Monte Carlo techniques and an Integrated Nested Laplace Approximation. On the municipality level and with environmental data at higher resolution, the study confirmed results of a previous publication, which had utilized exact locations of foxes and micro-habitat data. Furthermore, the preference of infected foxes for open landscapes with pasture was demonstrated in both regions despite different landscape characteristics. However, prediction of endemic areas was not possible alone on the basis of land-use classes in the study areas, as proximity of a naïve to an infected area, i.e. the probability of introduction of the parasite by neighbourhood, explained most of the variability in the spatial distribution of *E. multilocularis* in foxes.

3:15 PM - 3:30 PM (15 mins)

Predatory capacity of different copepods against *Aedes aegypti* larvae from Lahore

Nusrat Jahan*, and Nabila Kousar

Zoology Department, GC University Lahore 54000 Pakistan

Current study was evaluated for the biological control of *Aedes aegypti* larvae using *Mesocyclops* collected from different water bodies. The main objective was to control *A. aegypti* larvae using environment friendly method with no toxic effect on human and other animals. Single species culture of different species of *Mesocyclops* was prepared by using pure stock culture of *Chilomonas* and *Paramecium* in laboratory. Predatory capacities of the three identified species *M. aspericornis, M. pehpiensis* and *M. ogunnus* was evaluated in laboratory at different larval densities using different size containers. The mean predatory capacity/rate of female and male *M. aspericornis, M. pehpiensis* and *M. ogunnus* in 200 ml water was 83, 69 and 63% and 62, 60 and 50% at 10 larval densities post 24 hours exposure respectively. In general predatory capacity increased with decreased larval density. Moreover, predatory capacity of different species of *Mesocyclops* decreased with increasing volume and size of the containers against different larval density (25-250). In addition best predatory ratio was 1:20 with fixed number (100) of first instars in 200 ml container post 24 hours exposures. However, predatory capacity increased by increasing the number of copepods. Moreover, predatory capacity/rate of various *Mesocyclops* against 1st instars *Aedes* larvae decreased in the presence of alternative food. In general, the three identified species from Punjab have high potential for biological control of *Aedes* larvae. Overall *Mesocyclops aspericornis* is an effective predator as compared to the other two species for the control of dengue vector from Lahore, Pakistan.

Session F2 (RCJ) - One Health

Chair: Diana Williams, University of Liverpool

2:00 PM - 2:30 PM (30 mins)
One health: parasites and priorities
Paul Torgerson
Section of Epidemiology, Vetsuisse Faculty, University of Zurich, Switzerland.

One health is dedicated to improving the lives of all species and involves the integration of human medicine, veterinary medicine and environmental science. In 2005 it was estimated that of 1407 species of recognized human pathogens, 58% were zoonotic. Many emerging diseases for example, are parasitic zoonoses. Furthermore, an estimated 24% of the disease burden (healthy life years lost) and an estimated 23% of all deaths (premature mortality) was attributable to environmental factors, many of which promote the transmission of parasitic diseases. However, we live in a world of limited resources and hence it is essential to give priorities to control some diseases and perhaps neglect some others. The choice of which diseases to prioritize should be determined by the potential effectiveness of a disease control programme, its cost and the cost effectiveness or cost benefit of undertaking the intervention. Benefits not only include human health, but also animal health, environmental health and ecological health. Disease burden analysis is an important tool to support policy in determining which outcomes would be of most beneficial to society. Furthermore human disease burden is being increasingly measured by egalitarian non-financial measures which do not sit comfortably with measurements of economic costs in livestock. This adds additional challenges to assessing socio-economic burdens of zoonotic diseases. Using examples from the group of neglected zoonotic diseases, information regarding the socio-economic effects of these diseases are reviewed and the use of this information in decision making with regard to control and treatment of these diseases is discussed. A one health approach where the impact of diseases is considered across different sectors can be used to synergise control and improve cost effectiveness.

2:30 PM - 2:45 PM (15 mins)
Migration and the risk of animal trypanosomiasis on the Jos Plateau, Nigeria
Oluwashola Olaniyan, Picozzi,K.,MacLeod,E., Majekodunmi,AO., Igweh,AC., Dongkum,C., Lang,D. Welburn, SC.
Division of Pathway Medicine and Centre for Infectious Diseases, School of Biomedical Sciences, College of Medicine and Veterinary Medicine, The University of Edinburgh, Chancellor's Building, 49 Little France Crescent, Edinburgh EH16 4SB, UK

Until recently, the Jos Plateau in north central Nigeria was considered free of tsetse flies, vectors of human and animal trypanosomiasis in Africa. This disease severely limits livestock production in a large part of Africa but was of little relevance on the Plateau. Recent studies, however, show animal trypanosomiasis to be increasingly prevalent, leading to significant agricultural losses. Seasonal migration by Fulani pastoralists and their cattle is one of the reasons suggested for this increase. Once limited to the dry season (November-March), migration is now practiced during the wet season (April-October) as access to grazing and water is limited by expanding crop production on the Plateau. To improve our understanding of the epidemiology of the disease and design effective interventions, it is important to determine the risk that this practice poses to livestock on the Plateau In this work, a migrant herd was followed from June 2013 to February 2014. Captured tsetse flies and cattle blood samples were screened for trypanosomes using ITS-1 PCR. Results show that cattle infection was almost zero between June and September but rose to 16.7% (12/72, 95%CI 9.8-26.9%) by the end of November. The infection rate in tsetse was 21.3% (17/80, 95%CI 13.7-31.4%) with *Trypanosoma vivax* the predominant species in both cattle and flies. We conclude that animals moving through this route to the base of the Plateau are subject to a higher trypanosomiasis challenge than on the Plateau and may bring these infections back to the Plateau when they return.

2:45 PM - 3:00 PM (15 mins)

Arsenic, antimony and *Leishmania* - has arsenic contamination of drinking water in India led to treatment resistant kala-azar? *

Meghan Perry[1], Susan Wyllie[1], Vijay K Prajapati[3], Joris Menten[4], Andrea Raab[2], Joerg Feldmann[2], Dipankar Chakraborti[5], Shyam Sundar[3], Albert Picado[6], Marleen Boelaert[4], Alan Fairlamb[1]

[1.]Division of Biological Chemistry and Drug Discovery, College of Life Sciences, University of Dundee, Dundee DD1 5EH, Scotland, UK; [2.]College of Physical Sciences – Chemistry, Trace Element Speciation Laboratory, Meston Walk, University of Aberdeen, Aberdeen AB24 3UE, Scotland, UK; [3.]Infectious Disease Research Laboratory, Department of Medicine, Institute of Medical Sciences, Banaras Hindu University, Varanasi 221 005, India; [4.]Epidemiology & Disease Control Unit, Department of Public Health, Prince Leopold Institute of Tropical Medicine, Nationalestraat 155, B-2000 Antwerp, Belgium; [5.]School of Environmental Studies, Jadavpur University, Kolkata 700032, India; [7.]Barcelona Centre for International Health Research (CRESIB, Hospital Clínic-Universitat de Barcelona), Barcelona, Spain

In Bihar state, India, the cure rate of antimonial compounds (Pentostam) in the treatment of visceral leishmaniasis (VL) has declined from over 85% to less than 50%. This has been attributed to prolonged, widespread misuse of antimonials within the Indian private healthcare system. An alternative resistance hypothesis is that exposure to arsenic in drinking water in this region has resulted in antimony-resistant *Leishmania* parasites. *Leishmania donovani* were serially passaged in mice exposed to environmentally-relevant levels of arsenic in drinking water. Arsenic accumulation in organs of these mice was proportional to exposure. After five passages, isolated parasites were refractory to Pentostam in drug sensitivity assays. Treatment of infected mice with Pentostam confirmed that these parasites retained resistance *in vivo*, supporting this hypothesis. A retrospective field study on a cohort of antimony treated VL patients was performed in an arsenic contaminated area of Bihar to evaluate the presence of an increased risk of treatment failure and death in those exposed to arsenic. This demonstrated a significant increased risk of death from VL in arsenic exposed patients but did not indicate a significant relationship between arsenic exposure and antimonial treatment failure. Collectively these data suggest that it is biochemically possible that arsenic contamination may have contributed to the development of antimonial resistance in Bihar although issues of underpower and the retrospective nature of our epidemiological study made it difficult to conclusively demonstrate this. Further research into the relationships between arsenic exposure and antimonial treatment failure and death is required.

3:00 PM - 3:15 PM (15 mins)

WIPO Re:Search - A Catalyst for Success: Channeling the Expertise of Industry, Academic, and Nonprofit Organizations toward Neglected Disease Research

Katy Graef, Roopa Ramamoorthi, Jennifer Dent

BIO Ventures for Global Health 401 Terry Avenue North Seattle, Washington 98109 United States

Cross-sector collaborations are becoming increasingly integral to the discovery and development of biomedical products. Yet while many of these collaborations were established to encourage R&D for diseases such as cancer and cardiovascular indications, diseases of poverty – which affect over one billion individuals worldwide – have historically been overlooked by these collaborative efforts. WIPO Re:Search was established to address this disparity by connecting the intellectual property assets of pharmaceutical companies, such as compounds, data, and technical know-how, with academic and nonprofit neglected disease experts capable of utilizing these assets to advance their own product discovery efforts. WIPO Re:Search was founded by the World Intellectual Property Organization (WIPO), BIO Ventures for Global Health (BVGH), and leading pharmaceutical companies, academic institutions, and research organizations to promote cross-sector collaborations to accelerate the development of new drugs, vaccines, and diagnostics for neglected tropical diseases, malaria, and tuberculosis. As the Partnership Hub Administrator, BVGH works with researchers to understand their research and specific requests, identifies members able to fulfil those requests, and helps forge mutually beneficial collaborations with clearly defined roles, responsibilities, and expectations. In addition, BVGH proactively identifies and proposes novel collaboration opportunities to members. This proactive partnering has proven to be highly successful and is responsible for the 49 WIPO Re:Search arrangements facilitated to date – some of which have advanced from screens and identification of hits to dose response and optimization studies. This presentation will introduce WIPO Re:Search and describe the innovative research involving helminths, kinetoplastids, and malaria that it has facilitated.

Session A3 (WRL) - Kinetoplastida - RNA biology

Chair: C Clayton, University of Heidelberg

4:00 PM - 4:30 PM (30 mins)
Moving forward: from procyclics to metacyclics in *Trypanosoma brucei*
Elisabetta Ullu
Yale

The infectious metacyclic form of the human pathogen *Trypanosoma brucei* develops in the salivary glands of tsetse flies. Changes in gene expression underlying infectivity acquisition are not well defined. An *in vitro* metacyclogenesis system, based on overexpression of the RNA binding protein RBP6, allowed us to survey the proteome and transcriptome of metacyclics and revealed several features of this cell type and its adaptations in preparation to infect a mammalian host.

4:30 PM - 4:45 PM (15 mins
Purification of specific mRNPs via the nascent polypeptide
Diana Patricia Inchaustegui Gil, and Christine Clayton
Zentrum Molekular Biologie Heidelberg

Translating mRNAs are expected to associate with many RNA-binding proteins (RBPs). The interactions between these proteins and *cis*-regulatory motifs, present in the untranslated regions (UTRs) of the mRNAs, determine the cytoplasmic fate of mRNAs. To detect key factors involved in the regulation of gene expression in trypanosomes, a method to affinity purify specific ribosome-associated messenger ribonucleoprotein particles (mRNPs) was designed. The purification relies on three streptavidin-binding peptides (3SBP) at the N-terminus of the nascent polypeptide. These 3SBP connected to the actively translating mRNAs on polyribosomes will bind to the streptavidin matrix and the attached proteins eluted by boiling. The method was tested using a known RNA-protein interaction in trypanosomes. As reported previously [Droll, D., et al, PLoS Pathog, 2013], a zinc finger protein, ZC3H11 binds to an AU-rich element present in the HSP70 3' UTR. This interaction stabilises the mRNA. mRNPs were purified using our affinity purification method. Two independent purifications were made, one using the complete HSP70 3' UTR and another without the AU-rich element, as control. Indeed, ZC3H11 was detected in the purification when the AU-rich element was present in the HSP70 3' UTR, and was absent from the control purification. Other affinity purifications were done using an EP 3' UTR reporter. Mass spectrometry revealed translation factors, ribosomal proteins, proteins of the degradation machinery and other proteins associated with mRNPs. Interestingly, a candidate CCCH zinc-finger protein, that has not been detected before in other purifications, was found and is currently under characterisation.

4:45 PM - 5:00 PM (15 mins)
Comparative ribosome-profiling reveals extensive translational complexity in different *Trypanosoma brucei* life-cycle stages *
Juan José Vasquez, Chung-Chau Hon Ramona Derr Jens T. Vanselow Andreas Schlosser T. Nicolai Siegel
Juan-José Vasquez, Ramona Derr and T. Nicolai Siegel: Research Center for Infectious Diseases, University of Wuerzburg, Wuerzburg, 97080, Germany Chung-Chau Hon: Institut Pasteur, Unité Biologie Cellulaire du Parasitisme, Département Biologie cellulaire et infection, Paris, 75015, and INSERM U786, Paris, 75015, France Jens T. Vanselow and Andreas Schlosser: Rudolf Virchow Center, University of Wuerzburg, Wuerzburg, 97080, Germany

Gene expression is a fundamental and tightly controlled cellular process that is regulated at multiple steps including the transcriptional and post-transcriptional levels. However, due to their interdependence, the exact contributions of the individual steps to global gene expression remain unknown in any organism. The apparent absence of transcription initiation regulation for RNA pol II in *Trypanosoma brucei* should greatly simplify the task of elucidating the contribution of translation to global gene expression. Therefore, we have sequenced ribosome-protected mRNA fragments in *T. brucei*, permitting the genome-wide analysis of RNA translation and translational efficiency. We find that the latter varies greatly between life-cycle stages of the parasite and ~100-fold between genes, thus contributing to gene expression to a similar extent as RNA stability. The ability to map ribosome positions at sub-codon resolution revealed extensive translation from upstream ORFs (uORFs) located within 5´ UTRs. Using a luciferase–β-galactosidase double-reporter system we are currently evaluating the impact of uORFs and sequence motifs found in the 3´ UTR on translational

efficiency and RNA stability. Finally, profiling of ribosome positions enabled the identification of hundreds of previously un-annotated putative coding sequences (CDSs). Evaluation of existing proteomics and genome-wide RNAi data confirmed the translation of some of these previously un-annotated CDSs and suggested an important role for more than 200 of those CDSs in parasite survival, especially in the form that is infective to mammals. Overall our data show that translational control plays a prevalent and important role in different parasite life-cycle stages of *T. brucei*.

5:00 PM - 5:15 PM (15 mins)

Post-transcriptional control of mRNAs by ZC3H11 and MKT1 in *T. brucei*

Dorothea Droll, Igor Minia, Aditi Singh, Esteban Erben, Abeer Fadda, Christine Clayton
DKFZ- ZMBH Alliance, Universität Heidelberg, INF282, 69120 Heidelberg

In kinetoplastids, virtually all control of protein-coding gene expression is post-transcriptional, with mRNA degradation playing a central role. Unlike other eukaryotes, trypanosomes do not induce transcription of heat shock protein genes upon elevated temperatures. Instead, they selectively stabilize heat shock transcripts under stress conditions. We have found that the *Trypanosoma brucei* CCCH zinc finger protein ZC3H11 is essential for the trypanosome heat shock response. ZC3H11 binds to an AU-rich sequence element in the 3'UTRs of mRNAs that encode the major heat shock proteins HSP70 and HSP90 and various co-chaperones, and stabilizes them after heat shock. Expression of ZC3H11 is also induced upon heat shock: preliminary results indicate that both protein stability and translational control are important for this. We have also determined the mechanism by which ZC3H11 stabilises mRNAs in bloodstream forms. ZC3H11 interacts with trypanosome homologues of yeast MKT1 and PBP1. All three proteins are essential for bloodstream-form trypanosome survival and increase reporter expression in a tethering assay. Using a yeast two-hybrid screen and tandem affinity purification, we found that trypanosome MKT1 interacts with multiple RNA-binding proteins and other potential RNA regulators, placing it at the centre of a post-transcriptional regulatory network. Recruitment of MKT1-containing regulatory complexes to mRNAs via sequence-specific mRNA-binding proteins could thus control several different post-transcriptional regulons.

5:15 PM - 5:30 PM (15 mins)

Transcriptome-wide analysis of mRNA decay in trypanosomes reveals complex degradation kinetics and novel control mechanisms

Christine Clayton[1], Abeer Fadda[1], Mark Ryten[2], Dorothea Droll[1], Federico Rojas[3], Jurgen Haanstra[4], Barbara Bakker[4], Valentin Färber[1], Keith Matthews[3]
[1.]Zentrum für Molekulare Biologie der Universität Heidelberg (ZMBH), DKFZ-ZMBH Alliance, Im Neuenheimer Feld 282, 69120 Heidelberg, Germany; [2.]Paddington, England; [3.]Centre for Immunity, Infection and Evolution, Institute for Immunology and infection Research, School of Biological Sciences, Ashworth Laboratories, University of Edinburgh, West Mains Road, Edinburgh EH9 3JT, Scotland; [4.]University of Groningen, Hanzeplein 1, Postbus 30.001, 9700 RB Groningen, The Netherlands

African trypanosomes are uniquely suited to the study of post-transcriptional mRNA control mechanisms. Due to the lack of crontrol of transcription initiation, steady-state mRNA levels have to be determined by the rates of processing and turnover. We have measured the rates of *trans*-splicing and mRNA decay in two life cycle stages of trypanosomes, the bloodstream and the procyclic form, by transcription inhibition and RNASeq. Many trypanosome mRNAs are spliced within a few minutes of synthesis, and as expected, developmentally regulated mRNAs often show regulated decay rates. We found that mRNAs with short half-lives often show initial fast degradation, followed by a slower phase. They are targets of the 5'-3' exoribonuclease XRNA. For many longer-lived mRNAs the first step in degradation is removal of the poly(A) tail; the body of the mRNA is initially not degraded at all, but is later rapidly destroyed. Rates of mRNA decay are quite good predictors of steady state levels for short mRNAs. In contrast, mRNAs longer than 3 kb are generally less abundant than shorter ones and their decay rates predict their steady state levels poorly. A new mathematical model for gene expression can mimic these results and shows that regulation of RNA processing can only affect the mRNA level if processing is competing with co-transcriptional mRNA degradation. Modelling and results from RNAi also show that co-transcriptional degradation can account for the loss of long mRNAs.

Session B3 (WRR) - Apicomplexa - Biochemistry

Chair: Oliver Billker, Wellcome Trust Sanger Institute

4:00 PM - 4:30 PM (30 mins)

Using quantitative phosphoproteome and proteome analysis to identify signaling pathways controlled by parasitic kinases

Moritz Treeck[1,2], John Sanders[2], Matthew Child[2], Gustavo Arrizabalaga[3], Joshua Elias[2] & John Boothroyd[2]
[1.] National Institute of Medical Research, Mill Hill, UK; [2.] Stanford University, School of Medicine, Stanford University, USA; [3.] University of Indianapolis, School of Pharmacology, USA

Phosphorylation is one of the major means of transducing signals within a cell. Using near-system-wide quantitative phosphoproteome and proteome analysis, we are trying to identify signaling pathways in apicomplexan parasites like *Plasmodium falciparum* and *Toxoplasma gondii* that are essential for their parasitic lifestyle. For example, we have identified proteins in *Toxoplasma* that are differentially phosphorylated during induced rapid egress from the host cell in dependence of a calcium dependent kinase (TgCDPK3). Beyond the expected proteins involved in calcium-signaling and motility, our results helped us to identify a yet undefined, but novel role for the kinase in this parasites

4:30 PM - 4:45 PM (15 mins)

Comparative metabolomics of erythroid lineage: implications for malaria control *

Anubhav Srivastava[1],Darren Creek[2],Krystal Evans[3],Louis Schofield[3],Michael Barrett[1],Malcolm McConville[2],Andy Waters[3]
[1.] Wellcome Trust Centre for Molecular Parasitology, University of Glasgow. [2.] Department of Biochemistry and Molecular Biology, Bio21 Molecular Science and Biotechnology Institute, University of Melbourne, Australia. [3.] Walter and Eliza Hall Institute of Medical Research, Melbourne, Australia

Plasmodium spp. are the causative agents of malaria and are obligate intracellular parasites that inhabit circulating cells of the erythroid lineage during the blood stage pathogenic phase of their life cycle. Whereas *Plasmodium falciparum* can readily proliferate in mature erythrocytes (normocytes) the more widespread and economically more important *Plasmodium vivax* exhibits a strong preference for immature reticulocytes. The rodent model of malaria *Plasmodium berghei* has a similar preference for growth in reticulocytes prompting a comparison of the metabolomes of rodent and human normocytes and reticulocytes in the expectation that metabolomic differences might form a nutritional basis for the parasite preference. Reticulocytes were found to be metabolically enriched with some active metabolic pathways in common with those predicted to be present and active in the parasite itself. To test whether the reticulocyte metabolites are exploited for parasite survival and growth, attempts were made to disrupt genes encoding key enzymes of intermediary carbon metabolism and pyrimidine biosynthesis creating potential auxotrophs for these pathways. Both pathways could be disrupted and 4 different single gene deletion mutants were successfully generated. Three of the four mutants failed at different points to complete transmission in mosquitoes as a result of delayed auxotrophy once the reticulocyte is abandoned. The findings have potential implications for drug therapies against blood stage malaria that target parasite metabolism which should differ according to the differing target host blood cell distributions of human malaria.

4:45 PM - 5:00 PM (15 mins)

Polycistronic and antisense transcription of the *Plasmodium* apicoplast genome

Ellen Nisbet, Davy Kurniawan, Erin Butterfield, Harrison Bowers, Christoper Howe
Dept. Biochemistry University of Cambridge Tennis Court Rd Cambridge CB2 1QW

The *Plasmodium* apicoplast contains a circular, 35 kb genome encoding proteins, tRNA, SSU and LSU rRNA genes. Extensive transcript mapping across genome, using RT-PCR on both circular and non-circularized RNA has shown that all examined regions are transcribed. Many transcripts are polycistronic, and cover both protein coding genes and tRNA/rRNA genes. Although many transcripts end near tRNA genes, others appear to have no conserved starting or ending points. These transcripts are processed to mRNA molecules, corresponding to predicted genes. Differential cleavage of polycistronic transcripts allows the expression of overlapping genes. We have also identified polycistronic antisense transcripts, again covering all examined regions of the genome. Similar transcripts are found in photosynthetic chloroplast species, where they play a role in the regulation of chloroplast gene expression. We are examining the role of such transcripts in *Plasmodium.*

5:00 PM - 5:15 PM (15 mins)

Development of a directional, amplification-free RNA-seq protocol optimised for AT-rich *Plasmodium* parasites

Lia Chappell, Chris Newbold, Julian Rayner, Matt Berriman
Wellcome Trust Sanger Institute

High-throughput sequencing of cDNA libraries (RNA-seq) is a powerful tool that can be used to investigate gene expression on a global scale. However, standard protocols include a number of steps that are biased against AT-rich *Plasmodium* RNA (the AT-content is around 80% in *P. falciparum*), including PCR-based amplification of the final sequencing library. We have developed a directional, amplification-free RNA-seq library preparation protocol for the Illumina platform that can be used with *Plasmodium* parasites, which requires a similar input amount of total RNA as PCR-based protocols. This approach significantly decreases the biases observed in AT-rich regions of the transcriptome, resulting in much more even coverage and more accurate transcript levels- valuable for comparative gene expression analysis. This new approach also reveals extensive transcription within extremely AT-rich intergenic regions (AT-content as much as ~90%), which shows that the *Plasmodium* transcriptome extends significantly beyond the annotated protein coding regions.

5:15 PM - 5:30 PM (15 mins)

The antimalarial action of FK506, rapamycin and non-immunosuppressive congeners: evidence for a direct effect on FK506-binding protein

Bell, A[1], Monaghan P[1], Bianchin A[1,2], Shaw W[1], Wilkinson B[3,4], Chubb A[2], Shields D[2].
[1.] Dept. of Microbiology, School of Genetics & Microbiology, Moyne Institute, Trinity College Dublin, IRELAND [2.] Complex & Adaptive Systems Laboratory and School of Medicine & Medical Science, University College Dublin, IRELAND [3.] Isomerase Therapeutics Ltd., Chesterford Research Park, Cambridge, U.K. [4.] Dept. of Molecular Microbiology, John Innes Centre, Norwich, U.K.

FK506 and rapamycin are immunosuppressive drugs that act principally on T-lymphocytes. The receptors for both drugs are FK506-binding proteins (FKBPs) but the molecular mechanisms of immunosuppression differ. The FK506–FKBP complex inhibits the protein phosphatase calcineurin, blocking a key step in T-cell activation, while the rapamycin–FKBP complex binds to the protein kinase TOR, which is involved in a later signalling pathway. Both drugs, and certain non-immunosuppressive compounds related to FK506, have potent antimalarial activity. The mechanisms of antimalarial action of these inhibitors have however remained obscure. *Plasmodium* produces an FKBP (PfFKBP35) and a calcineurin. Given conflicting evidence on the involvement of this calcineurin in the action of FK506, and the absence of an apparent TOR homologue, we set out to establish whether inhibition of PfFKBP35 itself might be responsible for the antimalarial effects of FK506 and rapamycin. In the absence of knock-out or knock-down data, we propose that similarities in the antiparasitic actions of FK506 and rapamycin would constitute indirect evidence for this hypothesis. FK506 and rapamycin were compared for (i) susceptibility of different intra-erythrocytic stages, (ii) kinetics of parasite killing and (iii) pharmacodynamic interactions with other antimalarial agents. In all cases the two drugs had similar properties. Our data suggest that ligands binding selectively to PfFKBP35 may be promising antimalarial agents. In agreement with this idea, non-immunosuppressive rapamycin analogues had potent and selective antimalarial activity. One analogue was superior to rapamycin and all other macrolactone FKBP ligands reported to date.

Session C3 (WRA) - Kinetoplastida - Diseases: Epidemiology, Pathology, Diagnosis and Treatment III

Chair: Sanjeev Krishna, St George's, University of London

4:00 PM - 4:30 PM (30 mins)

Pathways to *Leishmania* persistence: in vivo veritas?

Paul Kaye, Paul Kaye
York University

Leishmania infection is characterized by long-term persistence, even in fully immunocompetent hosts. Yet how this is achieved at the intracellular, cellular and tissue level is not fully understood. Using *Leishmania donovani* infection of mice as a model system, we have been exploring how concepts that have arisen largely from in vitro studies play out in different tissue microenvironments. This presentation will summarize our recent studies, employing in vivo imaging and ex vivo analysis coupled with computational modeling, to evaluate i) the role of cell intrinsic and inflammation-driven pathways of innate resistance and ii) the extent to which parasites modify host cell signalling cascades to aid their persistence.

4:30 PM - 4:45 PM (15 mins)

Bioluminescence imaging of chronic *Trypanosoma cruzi* infections reveals tissue-specific parasite dynamics and heart disease in the absence of locally persistent infection

Amanda Fortes Francisco[1], Michael D. Lewis[1], Martin C. Taylor[1], Hollie Burrell-Saward[2], Alex P. McLatchie[1,3], Michael A. Miles[1] and John M. Kelly[1]
[1.] *Department of Pathogen Molecular Biology,* [2.] *Department of Immunology and Infection, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, Keppel Street, London, WC1E 7HT, United Kingdom* [3.] *Current address: Institute of Immunity and Transplantation, UCL Department of Immunology, Rowland Hill Street, London, NW3 2PF, United Kingdom*

Chronic *Trypanosoma cruzi* infections lead to cardiomyopathy in 20-30% of cases. A causal link between cardiac infection and pathology has been difficult to establish because of a lack of robust methods to detect scarce, focally distributed parasites within tissues. We developed a highly sensitive bioluminescence imaging system based on *T. cruzi* strain CL Brener expressing a novel luciferase that emits tissue-penetrating orange-red light. This enabled long-term serial evaluation of parasite burdens in individual mice with an *in vivo* limit of detection of significantly less than 1000 parasites. Parasite distributions during chronic infections in BALB/c mice were highly focal and spatiotemporally dynamic, but did not localize to the heart. End-point *ex vivo* bioluminescence imaging allowed tissue-specific quantification of parasite loads with minimal sampling bias. During chronic infections, the gastro-intestinal tract, specifically the colon and stomach, was the only site where *T. cruzi* infection was consistently observed. Quantitative PCR-inferred parasite loads correlated with *ex vivo* bioluminescence and confirmed the gut as the parasite reservoir. Chronically infected mice developed myocarditis and cardiac fibrosis, despite the absence of locally-persistent parasites. These mice also displayed a qualitative increase in the thickness of the smooth muscle layer of both the colon and the oesophagus, combined with an increased lumen circumference. Inflammation and goblet cell hyperplasia/hypertrophy were observed as progressive phenomena. In summary, our data identify the gut as a permissive niche for long-term *T. cruzi* infection and show that canonical features of Chagas disease can occur without continual myocardium-specific infection.

4:45 PM - 5:00 PM (15 mins)

Aquaporin 2 is the main determinant for pentamidine and melaminophenyl arsenical resistance in *Trypanosoma brucei* spp.

Harry De Koning[1], Jane C. Munday[1], Anthonius A. Eze[1,2], Nicola Baker[3], Lucy Glover[4], Fabrice E. Graf[4] Pascal Mäser[5], David Horn[4]
[1.] *Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, United Kingdom.* [2.] *Department of Medical Biochemistry, University of Nigeria, Enugu Campus, Enugu, Nigeria.* [3.] *University of Dundee, Scotland, UK.* [4.] *Swiss Tropical and Public Health Institute, Basel, Switzerland*

It has been known for decades that resistance to melaminophenyl arsenicals in trypanosomes is often linked to reduced sensitivity to diamidines, particularly pentamidine. The identification of the P2/TbAT1 aminopurine transporter as a carrier for these agents and for the related veterinary diamidine diminazene appeared to settle the resistance mechanism. However, loss of TbAT1 is principally associated with diminazene resistance but melarsoprol/pentamidine cross-resistance (MPXR) is the result of loss of the High Affinity Pentamidine Transporter (HAPT1). The genetic identity of HAPT1 remained unknown until a genome-wide RNAi library screen found that knockdown of an aquaporin could lead to MPXR. Follow-on with double knockouts and re-expression identified TbAQP2 as the main determinant of MPXR, while its expression did not affect diminazene sensitivity. TbAQP2 was found to be absent or changed in all MPXR lab strains examined, whether adapted *in vivo* or *in vitro*, to arsenicals or pentamidine, and from *T. b. brucei, T. b. gambiense* and *T. b. rhodesiense* alike, whereas WT-AQP2 was found in the isogenic, sensitive reference strains. Expression of TbAQP2 in even the most resistant strain completely reversed MPXR, and re-introduced HAPT1 function and transport kinetics. Indeed, expression of TbAQP2 in *Leishmania mexicana* dramatically sensitised promastigotes to cymelarsan and pentamidine, introducing a pentamidine transport activity indistinguishable from HAPT1. We conclude that TbAQP2 functions not just as a classical aquaglyceroporin but also as a high affinity drug transporter. This is clinically relevant as clinical outcomes of melarsoprol treatment correlated to the presence of the trypanosomes containing a wild-type AQP2 allele.

5:00 PM - 5:15 PM (15 mins)

Host immunity is regulated by alternatively activated macrophages in Indian Post Kala-Azar Dermal Leishmaniasis

Mitali Chatterjee[1], Debanjan Mukhopadhyay[1], Shibabrata Mukherjee[1], Nilay K. Das[2]

[1.] Dept of Pharmacology, Institute of Postgraduate Medical Education & Research, 244B, Acharya JC Bose Road, Kolkata, West Bengal, INDIA and [2.] Dept. of Dermatology, Calcutta Medical College, Kolkata, West Bengal, INDIA

Amongst the diverse manifestations of Leishmaniasis, Post Kala-azar Dermal Leishmaniasis (PKDL), caused by *Leishmania donovani* is possibly the most intriguing as it generally develops after apparent cure from Visceral Leishmaniasis (VL). In view of its pivotal role in fuelling the transmission of VL, the immune status of monocytes-macrophages, essential host cells for the *Leishmania* parasite was evaluated in Indian PKDL (n = 25). Intramonocytic levels of pro-inflammatory (IL-6, IL-8, IL-1β, TNF-α) and anti-inflammatory cytokines (IL-10, TGF-β) were measured along with their redox status (levels of nitric oxide (NO), reactive oxygen species (ROS) and non protein thiols). Additionally, the expression of TLR-2, TLR-4 and phenotypic markers (CD14/CD16, CD40, CD80, CD86, HLA-DR, CD54) were evaluated in circulating monocytes. Furthermore, the activation status of monocytes-macrophages was measured in circulation and dermal lesions in terms of their expression of the mannose receptor, arginase-I and the Vitamin D-signaling pathway. In PKDL, monocytes showed a decreased expression of CD16, CD86, TLR-2 and TLR-4 along with decreased levels of NO, ROS and pro-inflammatory cytokines (IL-6, IL-8, IL-1β, TNF-α). Furthermore, their expression of co-stimulatory molecules (CD40, CD86) was down-regulated as was HLA-DR. Within monocytes, levels of thiols, IL-10 and TGF-β were raised concomitant with altered vitamin D-signaling. Macrophage polarization (M2 phenotype) was corroborated by an increased expression of mannose receptor and arginase-I in systemic monocytes and tissue macrophages. Taken together, in PKDL, altered immune functions caused monocytes to be polarized towards alternative activation (M2), which following anti-leishmanial chemotherapy were repolarized towards a classically activated M1 form.

5:15 PM - 5:30 PM (15 mins)

***Leishmania mexicana* modulates dendritic cell migration towards draining lymph nodes ***

Jenny Crowe, Gareth Westrop, Jeremy Mottram, James Alexander and Owain Millington

Centre for Biophotonics, Strathclyde Institute of Pharmacy & Biomedical Sciences, University of Strathclyde, Glasgow, G4 0RE. GBRC, University Place, University of Glasgow, Glasgow, G128TA.

Migration of dendritic cells (DCs) is a critical step in the activation of an adaptive immune response. Expression of the chemokine receptor CCR7 facilitates DC migration towards CCL19-expressing lymphatic vessels and the draining lymph node. Consequently, parasite manipulation of chemokines and chemokine receptors presents a potential mechanism for evasion of protective immunity. Here, we examined the impact of *Leishmania mexicana* on DC migration. DCs co-cultured with *L. mexicana* promastigotes fail to express CCR7, are refractory to LPS-stimulation and show reduced migration towards CCL19 *in vitro*. Furthermore, *in vivo* DCs exposed to *L. mexicana* fail to infiltrate into lymphatic vessels. Further studies provide evidence demonstrating that the mechanism of immune-evasion is closely associated with the parasite virulence factor cysteine protease B (CPB). Our data demonstrate that *L. mexicana* can impair immune activation and that perturbation of DC migration plays a key role in this process. Identifying CPB as an important virulence factor that influences DC migration may provide a potential therapeutic target for preventing immune-evasion by *Leishmania*.

Session D3 (RCA) - Helminths - Human immunology and morbidity

Chair: Shona Wilson, University of Cambridge

4:30 PM - 4:45 PM (15 mins)

Identifying and quantifying morbidity markers associated with *Schistosoma haematobium* infection in children *

Welcome Mkululi Wami[1],Norman Nausch[1], Nicholas Midzi[2] Takafira Mduluza[3], Mark Woolhouse[1+], Francisca Mutapi[1]

[1.] Institute of Immunology & Infection Research, [1+.] Centre for Immunity, Infection & Evolution, Ashworth Laboratories, University of Edinburgh, West Mains Road, Edinburgh EH9 3JT, UK; [2.] National Institute of Health Research, PO Box CY 573, Harare, Zimbabwe and Research Council of Zimbabwe, Mount Pleasant Business Park, Harare, Zimbabwe; [3.] Department of Biochemistry, University of Zimbabwe, PO Box 167, Harare, Zimbabwe

Urogenital schistosomiasis, caused by *Schistosoma haematobium* is highly prevalent in sub-Saharan Africa. Our studies and those of others have shown significant levels of infection in preschool and primary school-aged children, however, there still remains a gap in knowledge about associated morbidity in the younger age group. Schistosomiasis symptoms are nonspecific, thereby complicating morbidity diagnosis. The aim of this study was to identify schistosomiasis-related morbidity markers and quantify the levels of morbidity attributable to schistosome infection in children. The study was conducted in Murewa district, Zimbabwe, a high *S. haematobium* transmission area. Morbidity was determined in children aged 1-10 years using dipsticks, urine albumin-creatinine ratio (UACR), questionnaires and clinical examination. Infection was determined using parasitology and serology. Non-metric multidimensional scaling was used to simultaneously investigate differences in several morbidity markers measured using dipsticks between children. The effects of age and sex on morbidity prevalence were investigated using logistic regressions. Dipstick haematuria and proteinuria were identified as responsible for most differences in dipstick morbidity markers observed in this population. The associations between infection and morbidity prevalence were found to be age-dependent, but not sex dependent. Furthermore, 68.8% (95% CI: 54.8-78.5%) of high abnormal UACR was attributable to some children being *S. haematobium* infection positive based on parasitology. Compared to other techniques, questionnaires and physical examinations were less able to identify morbidity in this study population. UACR, dipstick haematuria and proteinuria are important schistosomiasis-related morbidity markers in children. These findings are important for the planning of control programs.

4:45 PM - 5:00 PM (15 mins)

Anti-glycan antibody responses upon infection or vaccination with *Schistosoma mansoni*

Angela van Diepen[1], D. Linh Nguyen[1], Arend-Jan van der Plas[1], Nahome Kribbe[1], R. Alan Wilson[2], David W. Dunne[3], and Cornelis H. Hokke[1]

[1.] Department of Parasitology, Leiden University Medical Center, Leiden, The Netherlands; [2.] Centre for Immunology & Infection, Department of Biology, University of York, York, United Kingdom; [3.] Department of Pathology, University of Cambridge, Cambridge, United Kingdom

Schistosomiasis is a chronic and potentially deadly parasitic disease that affects millions of people in (sub)tropical areas. Immunity to Schistosoma can be acquired, but this takes many years of exposure, multiple infections and treatments, and maturation of the immune system. Most antibodies generated are directed against the numerous schistosome glycans, but the precise structure of the glycan antigens and the relation to immunity are poorly understood. Anti-glycan antibodies can be studied efficiently using glycan microarrays. We have generated a microarray containing hundreds of naturally occurring glycans isolated from different life stages of *S. mansoni*. To study the specificity and nature of anti-glycan antibodies in schistosomiasis, we have applied this microarray to the analysis of anti-glycan IgG and IgM in sera from different age groups within the *S. mansoni*-endemic Musoli community in Uganda. We observed age-dependent differences in responses, especially when looking at changes before and after treatment with PZQ. Also within age groups we observed differences between groups with high and low infection intensities. In addition we have used sera from baboons vaccinated with irradiated *S. mansoni* cercariae to study longitudinally the development of anti-glycan responses and show that responses against cercarial lipid-derived glycans as well as cercarial O-glycans are strongly but gradually induced by repeated vaccination. Detailed results will be presented. Shotgun glycan microarrays allow the definition of groups of schistosome-infected individuals as well as glycan element clusters to which antibody responses are generated in different cohorts and settings.

5:00 PM - 5:15 PM (15 mins)

IgE responses to abundant antigens in the parasite *Schistosoma mansoni*: A link between allergy and the evolved immune response to metazoan parasites?

Edward Farnell[1], Colin Fitzsimmons[1], Frances Jones[1], Jakub Wawrzyniak[1], Edridah Tukahebwa[2] and David Dunne[1]

[1.] Department of Pathology, University of Cambridge, UK; [2.] Vector Control Division, Ugandan Ministry of Health, Uganda

It has long been accepted that IgE-mediated immune responses are involved in both allergic reactions and the evolved immune responses against metazoan parasites. It has been recently observed that many allergens and parasite IgE-binding antigens share similar structures and functions, however the mechanisms determining which antigens or allergens are targets of the IgE response remain unknown. We hypothesise that allergens and parasite IgE-binding antigens have similar structures, with a common pathway of immune recognition and that allergy is therefore the unregulated by-product of the evolved immune responses against metazoan parasites. In this study we used a bioinformatic approach to predict allergen-like proteins in a sample of abundant *Schistosoma mansoni mansoni* proteins using known allergen structures. These antigens were produced as recombinant proteins and serological responses, including IgG1, IgG4 and IgE from individuals in a population endemically infected with *S. mansoni*, were measured. These responses were then modelled alongside common demographic confounders to identify any antigens associated with increased or decreased post-treatment re-infection. We found not only that known allergen structures can be used to predict IgE-binding in antigens from *S. mansoni,* but also that some of the IgE-binding targets in *S. mansoni* are associated with IgE mediated protection against re-infection in individuals living in an area of high *S. mansoni* transmission. These results provide further evidence for our hypothesis, providing a link between known allergen structures, IgE-binding parasite antigens and the evolved protective immune response against metazoan parasites.

5:15 PM - 5:30 PM (15 mins)

High throughput-compatible identification of novel helminth allergens using a humanised basophil reporter cell line

Franco Falcone[1], Daniel Wan[1,2] Fernanda Ludolf-Ribeiro[3], Daniel Alanine[1,2], Owen Stretton[1], Eman Ali Ali[1], Nafal Al-Barwary[1], Xiaowei Wang[2], Adriano Mari[4, 5], Colin Fitzsimmons[6], David W. Dunne[6], Ryosuke Nakamura[7], Guilherme Oliveira[3], Marcos J. C. Alcocer[2]

[1.] Division of Molecular and Cellular Science, School of Pharmacy, University of Nottingham, NG7 2RD UK; [2.] School of Biosciences, University of Nottingham, Sutton Bonington Campus, Loughborough, LE12 5RD, UK; [3.] Genomics and Computational Biology Group, Centro de Pesquisas René Rachou, Fundação Oswaldo Cruz - FIOCRUZ, Belo Horizonte, MG, Brazil; [4.] Center for Molecular Allergology, IDI-IRCCS, Rome, Italy, and [5.] Associated Centres for Molecular Allergology, Rome, Italy; [6.] Department of Pathology, University of Cambridge, Tennis Road, Cambridge CB2 1QP; [7.] National Institute of Health Sciences, Tokyo, Japan

Human infection with *Schistosoma mansoni* is known to correlate with a progressive increase of serum IgE levels. It is also known that increasing levels of parasite-specific IgE are associated with resistance to infection or post-treatment re-infection. Thus the molecular targets of the IgE response are *a priori* very promising vaccine candidates. There is however a major unsolved conundrum specific to the development of anti-helminthic vaccines, as vaccinating with an allergen bears the inherent risk of potentially inducing hazardous allergic reactions in sensitised individuals. We have previously shown that human basophils are sensitised within 6 weeks of a single, low-dose infection with *Necator americanus* infective stage larva. Basophil activation could be detected by flow cytometry in the absence of measurable parasite-specific IgE levels in the serum. This suggests that basophils may offer a more sensitive way of measuring the presence of parasite antigen-specific IgE in infected individuals, and, perhaps more importantly in the context of vaccination, to demonstrate the ability of allergens to induce basophil or mast cell activation, in contrast to measuring allergen binding by specific IgE alone. Here, we demonstrate proof-of-principle of how humanised rat basophil leukaemia (RBL) cell lines, expressing the human high affinity IgE receptor and stably transfected with different reporter genes, in combination with a cell-free *in vitro* translation system and a set of stringent quality controls, can be used for assessment of allergenicity of *S. mansoni* antigens. This technology paves the way for high-throughput, genome-wide assessment of *S. mansoni* antigen allergenicity - the Schistosoma Allergome.

Session E3 (RCU) - Ecology - Disease transmission

Chair: Sarah Perkins, Cardiff University

4:00 PM - 4:30 PM (30 mins)

Genome analysis of clonally transmissible cancers in dogs and Tasmanian devils

Elizabeth Murchison

University of Cambridge, Department of Veterinary Medicine, Cambridge, United Kingdom and Wellcome Trust Sanger Institute, Cancer Genetics and Genomics, Hinxton, United Kingdom

Tasmanian devil facial tumour disease (DFTD) and canine transmissible venereal tumour (CTVT) are the only two known naturally occurring clonally transmissible cancers. These are cancers that are transmitted between individuals by the physical transfer of living cancer cells. Thus DFTD and CTVT are long-lived somatic cell lineages that each first originated once as cancers in single individuals but that have now spread through their respective host populations as parasitic clonal cell lineages. DFTD is spread by biting and is threatening its host species, the Tasmanian devil, with extinction. CTVT is a sexually transmitted cancer that affects dogs and has spread around the world together with its host. The genome sequences of DFTD and CTVT have revealed features of the individuals that first spawned these two lineages as well as patterns of mutation and selection that have driven the evolution and characterised the emergence and spread of these two unusual long-lived and divergent cancers. Clonally transmissible cancers are a poorly understood type of infectious pathogen; although there are only two known diseases of this type, such diseases can emerge rapidly and have disastrous implications for species conservation.

4:30 PM - 4:45 PM (15 mins)

A not so slow boat to China: an update on the story of *Biomphalaria*, invasive carriers of neotropical schistosomiasis, in China

Stephen Attwood

State Key Laboratory of Biotherapy, 1 Ke Yuan 4 Lu, Chengdu, Sichuan, China PR, 610041

In 1973 planorbid snails, then identified as *Biomphalaria straminea*, were discovered in Hong Kong, China. It was assumed that these snails had been introduced to Hong Kong via the import of tropical fish by air from South America. In 2012 *Biomphalaria* were found for the first time in Guangdong Province China. In view of the renewed interest in these invasive snails, a morphological and DNA-sequence based phylogenetic study was undertaken for seven populations of *Biomphalaria* snails collected in Guangdong. This talk discusses the species which may be present and the sources, routes and means by which these snails may have been introduced to China. Recent evidence suggests that the snails in Guangdong are the result of an independent invasion (probably several) and not of dispersal of the Hong Kong populations into mainland China; this and its implications will be discussed. Finally, the risk of these snails to public health in China is assessed. Reports in the scientific literature are currently expressing the view that the risk is mainly from the immigration of migrant workers; however, the findings of our study suggest that this risk is minimal. Nevertheless, there is a potential risk of sporadic localized outbreaks of schistosomiasis through introduction of *Schistosoma mansoni* in the intramolluscan stage and this will be discussed.

4:45 PM - 5:00 PM (15 mins)

Epidemiology of Human Leptospirosis in Malaysia (2004 -2012)

Mohd Zain Siti Nursheena, Benacer Douadi, Siti Nursheena Mohd Zain, Khebir,Verasahib and Thong Kwai Lin

Institute of Biological Science, Faculty of Science, University of Malaya, 50603, Kuala Lumpur. Malaysia; Disease Control Division, Ministry of Health Malaysia, Level 3, Block E10, Complex E, Precint 1, Federal Government Administrative Centre, 62590 Putrajaya, Malaysia

Leptospirosis is a globally important zoonotic disease caused by spirochetes of the genus Leptospira. The bacteria colonize the kidneys of reservoir animals and excreted via urine into the environment. Transmission to humans occurs either directly from exposure to contaminated urine or infected tissues, or indirectly via contact with contaminated soil or water. In Malaysia, there is limited information on the human cases in relation to incidence and distribution patterns. Therefore, the objective of this study was to review the epidemiology of human leptospirosis cases in Malaysia. Data obtained from all reported cases from the Malaysian Ministry of Health Malaysia public hospitals between 2004 and the

end of 2012 were analyzed and mapped using Geographic Information System (GIS) to determine risk factors associated with this illness. A total of 12,335 cases were reported between 2004 and 2012 with annual incidence rate ranged from 1.05 to 12.49 per 100,000 population and the case fatality rate (CFR) decreased from 6.05% in 2004 to 1.30% in 2012. The highest reported cases were from the west coast states of Peninsular Malaysia with most cases coinciding with the monsoon season. More males between 20−50 years old were most susceptible and most common amongst the Malays. In conclusion, due to the endemicity and emergence of leptospirosis in Malaysia, as well as the dramatic increase in reported cases over the last decade, there is a critical need of collaboration in strategies for control and prevention of this disease in order to reduce the risk of the infection.

5:00 PM - 5:15 PM (15 mins)

The consequences of coinfections for parasite transmission in in the mosquito *Aedes aegypti*

Alison Duncan, Philip Agnew, Valérie Noel, Yannis Michalakis
MIVEGEC, UMR CNRS-IRD-UM1-UM2 5290 Centre IRD 911 avenue Agropolis 34394 Montpellier CEDEX 5 France

The abiotic environment frequently determines host life history traits, however, investigation about how it affects parasite fitness in a co-infection is lacking. We investigate how co-infection of the mosquito *Aedes aegypti* with two microsporidian parasites (*Vavraia culicis* and *Edhazardia aedis*) influences the transmission opportunities for each parasite, directly, and indirectly through their affects on host traits. In a laboratory infection experiment we compared how co-infection, at low and high larval food, affected the probability of infection, within-host growth and transmission potential, for each parasite, compared to single infections. We also compared the total input number of spores used to seed infections with output number, in single and coinfections for each parasite. The effects of co-infection on parasite fitness were complex, especially for *V. culicis*. At low food co-infection reduced the chances of mosquitoes emerging as adults, thus increasing opportunities for *V. culicis*' horizontal, within-site transmission. However, co-infection reduced larval longevity and time available for *V. culicis* spore production. Altogether there was a negative net effect of co-infection on *V. culicis* whereby the number of spores produced was less than the number used to seed infection. Co-infections also negatively affected horizontal transmission of the more virulent parasite, *E. aedis*, through reduced longevity of pre-adult hosts. However, its potential transmission suffered less relative to *V. culicis*. Our results show abiotic environmental conditions influence co-infections via modification of both host and parasite life-history traits. The resulting interactions can strongly impact the transmission potential of co-infecting parasite species

5:15 PM - 5:30 PM (15 mins)

Transmission dynamics of pathogenic bacteria by free-living nematodes

Anaid Diaz, Olivier Restif
Department of Veterinary Medicine, Disease Dynamics Unit, Madingley Road, Cambridge CB3 0ES

Many opportunistic pathogens persist by establishing symbiotic associations with other species. For example, filarial nematodes carry symbiotic bacteria that cause river blindness disease in humans. Bacterial symbiotic interactions are not limited to parasitic worms, recent studies have suggested that free-living nematodes act as a reservoir of pathogens such as *Salmonella enterica* in the soil. Despite its potential importance, we know little about the natural interactions between free-living nematodes and bacteria. In particular, about the dynamics of bacteria colonisation inside worms or how bacteria are transmitted between worms. To address this, we are developing an experimental system to study the within and between-host dynamics of bacteria in free-living nematode populations. We are employing *Caenorhabditis elegans* as a host and three human pathogens as symbionts: *Escherichia coli, S. enterica Typhimurium* and *Pseudomonas aeruginosa*. By using microbiological and molecular techniques, we monitor bacterial numbers inside worms and in the environment. When assessing the within-worm bacterial dynamics, we have found that bacteria showed a rapid turnover and replacement. Our results further demonstrated that worms increased the spread of bacteria around the environment. This consequently increased the between-worm transmission in naïve worms. Altogether, we have successfully developed a system to study how bacteria use nematodes to persist and what factors can affect this. This system can be used to further understand how worms affect the evolution of pathogenic bacteria in response to stressful conditions.

Session F3 (RCJ) - Parasite epidemiology

Chair: Paul Torgerson, University of Zurich

4:00 PM – 4:30 PM (30 mins)
Climate change and the epidemiology of nematode parasites: issues of scale
Eric Morgan
Royal Veterinary College

In attempting to predict the effects of climate and other environmental change on parasite epidemiology, decisions must be made on the spatial and temporal scales to be addressed. Focusing on gastrointestinal nematodes of wild and domestic ungulates (e.g. *Nematodirus battus*), and lungworms of carnivores (*Angiostrongylus vasorum*), I consider how, in order to understand and adapt to these changes, we must appreciate the links between epidemiology and parasite life history, and their evolutionary context. Thus, the challenges faced by parasites infecting wild animals moulded their transmission strategies, and equipped them for success in domestic situations. This includes an ability to adapt rapidly to climate change. Therefore I argue that epidemiological predictions under climate change scenarios should take account also of evolutionary time scales. Since micro-evolution can lead to spatial heterogeneity in transmission patterns, the spatial scale at which predictions are made must also take account of local adaptation. This problem is explored with decision support systems for domestic animal keepers in mind.

4:30 PM - 4:45 PM (15 mins)
Immuno-epidemiological models predict novel markers for parasite resistance *
Joaquín Prada Jiménez de Cisneros, Michael J. Stear, Colette Mair, Thorsten Stefan, Louise Matthews
University of Glasgow

Gastrointestinal nematodes are a global cause of disease and death in humans, wildlife and livestock. Livestock infection has historically been controlled with anthelminthic drugs, but the development of resistance means that alternative controls are needed. The most promising alternatives are vaccination, nutritional supplementation and selective breeding, all of which act by enhancing the immune response. Currently, control planning is hampered by reliance on the faecal egg count (FEC), which suffers from low accuracy and a nonlinear and indirect relationship with infection intensity and host immune responses. We address this gap by using extensive parasitological, immunological and genetic data on the sheep – *T. circumcincta* interaction to create an immunologically explicit model of infection dynamics in a sheep flock that links host genetic variation with variation in the two key immune responses to predict the observed parasitological measures. Using our model, we show that the immune responses are highly heritable and by comparing selective breeding based on low faecal egg counts versus high plasma IgA responses we show that the immune markers are a much-improved measure of host resistance. In summary, we have developed a genetically and immunologically explicit model of host-parasite infections, and show that by integrating genetic, immunological and parasitological understanding we can identify new and more powerful markers for diagnosis and control.

4:45 PM - 5:00 PM (15 mins)
Transmission dynamics and control of *Fasciola hepatica* within sheep in the UK: the impact of population structure
Nicola Beesley[1], K. Cwiklinski[1], D. J. L. Williams[1], S. Paterson[2], J. E. Hodgkinson[1]
[1.] Veterinary Parasitology, Institute of Infection and Global Health, University of Liverpool [2.] Centre for Genomic Research, University of Liverpool

Fasciola hepatica is increasingly a cause of parasitic disease of economic and welfare importance in sheep and cattle in the UK. Understanding population structure is key to exploring the transmission dynamics and effective control of F. hepatica, in particular, how drug resistance genes develop and spread. We have validated 15 polymorphic microsatellites using a multiplex PCR/ capillary sequencing approach and genotyped 720 *F. hepatica* adults from sheep (n=20) in Scotland, England and Wales. Six loci showed evidence of null alleles. The nine remaining loci showed alleles (including private alleles) ranging from 2 to 30, with between 3 and 125 genotypes reported. The average heterozygosity was 0.772 with SD±0.127 in individual parasites. Significant differences in heterozygosity were found between 17 pairs of loci (ANOVA:P=0.000). Parasites with the same multilocus genotype [MLG] (clones) were seen in 17 of the 20 livers. A total of 634 distinct MLGs were found, giving a clonal proportion of 0.19 parasites, and a genotypic diversity of 0.880 (range

0.343 – 1/liver). All Psex values were significant (P<0.01). The FST value across all loci and livers was 0.0176 which suggests little genetic differentiation. In conclusion the majority of livers contain clones, with 51 MLGs observed multiple times, suggesting that following clonal expansion of the parasite within the snail intermediate host, metacercariae with the same MLG aggregate on pasture and are ingested by a sheep. The FST value suggests high gene flow within *F. hepatica* populations in the UK, so drug resistance alleles have the potential to spread quickly.

5:00 PM - 5:15 PM (15 mins)

Quantifying the effects of individual animal characteristics and climatological factors on faecal worm egg count shedding in donkeys

Christopher Corbett[1], S. Love[1], G.T. Innocent[2], I. J. McKendrick[2], J.B. Matthews[3], L. Matthews[1], M.J. Denwood[1], F.A. Burden[4]

[1.]College of Medical, Veterinary and Life Sciences, University of Glasgow, UK; [2.]Biomathematics & Statistics Scotland (BioSS), The King's Buildings, Edinburgh, UK; [3.]Moredun Research Institute, Midlothian, EH26 0PZ, UK; [4.]The Donkey Sanctuary, Sidmouth, Devon, EX10 0NU, UK

Cyathostomins, the predominant parasitic nematodes of equids, have developed varying degrees of resistance to all three classes of anthelmintic licensed for use in horses. It is essential that the effectiveness of alternative methods of control for these pathogens are quantified, including incorporating climatic data and the commonly advocated practice of removal of faeces from pasture. Here, we obtained monthly faecal worm egg counts (FWEC, n=4,460 individual counts) from 803 donkeys based at The Donkey Sanctuary (Devon, UK). The dataset also included age, sex, field, FWEC history and previous anthelmintic administrations in each individual, as well as the pasture hygiene management method applied in the field where the donkey was grazed. FWEC were analysed alongside local climatic data using a generalised linear mixed model to assess associations between these variables and each observed monthly FWEC. The preferred model was identified using a model selection algorithm based on penalised likelihoods, and associated a 2.1% decrease in FWEC per day with air frost two calendar months ago (p<0.001) and a 38% lower FWEC in groups with twice weekly manual faecal removal compared to those with no faecal removal (p=0.004). Other weather effects, both alone and as interaction terms with the average FWEC of the field were included in the model, alongside individual FWEC history with anthelmintic administration as interaction terms and date as a single term. Our study identifies factors that may be useful as part of on-going predictive modelling based methods of improving targeted selective therapy.

5:15 PM - 5:30 PM (15 mins)

Tick borne pathogens in Nigerian livestock; the influence of acaricide treatment on tick burden *

Joe Farrimond, Vincenzo Lorusso, Ayodele Majekodunmi, Charles Dongkum, Gyang Balak, Augustine Igweh, Sue Welburn, Kim Picozzi

Division of Pathway Medicine, Edinburgh University Medical School, The Chancellor's Building, 49 Little France Crescent, Edinburgh EH16 4SB, UK

Ticks and tick-borne pathogens represent a constant threat to cattle fitness and production throughout sub-Saharan Africa. To assess the risk associated with high parasite burden, ticks were collected in October 2010 from indigenous (*Bos indicus*) cattle in Plateau State, Nigeria and preserved in 70% ethanol before being morphologically identified. After DNA extraction, samples were subjected to a molecular protocol consisting of three simultaneous polymerase chain reactions (PCRs) followed by reverse line blot (RLB) hybridisation of PCR products. 272 adult Ixodid ticks were identified, including 87 *Rhipicephalus (Boophilus) decoloratus*, 104 *Rhipicephalus (Boophilus) annulatus*, 12 *Rhipicephalus (Boophilus) geigyi*, 34 *Hyalomma truncatum* and 35 *Amblyomma variegatum*. Pathogens detected included *Anaplasma marginale, A. bovis, A. centrale, Ehrlichia canis, E. ruminantium, E. sp. Omatjenne, Theileria mutans* and *Babesia bigemina*. A high frequency of co-infection with both *A. marginale* and *Rickettsia spp*. was also seen upon RLB hybridisation. Amongst those ticks that tested positive for *Rickettsia spp*., at least 15% have also tested positive for spotted-fever group rickettsiae. As part of a large scale BBSRC funded animal health project, half body counts were made to assess the influence of a pour-on acaracide. Observations were recorded over an 18 month period, with both counts and treatments being applied on a tri-monthly basis. This presentation discusses the consequences of high tick burdens on cattle populations maintained within the study site of the Jos Plateau, Central Nigeria; including a description of the genetic evidence of haematoparasite infection and the impact of chemical deterrents on tick burdens experienced by these animals.

Day 2 - 08/04/2014

Session A4 (WRA) - Kinetoplastida - Growth, form and metabolism I

Chair: Eva Gluenz, University of Oxford

11:00 AM - 11:30 AM (30 mins)
Expression site attenuation and development in *Trypanosoma brucei*
Markus Engstler, Christopher Batram, Nicola G. Jones, Christian J. Janzen, Sebastian M. Markert
Universität Würzburg

African trypanosomes possess hundreds of variant surface glycoprotein (VSG) genes, but only one is expressed from a telomeric expression site (ES) at any given time. We found that the expression of a second VSG alone is sufficient to repress the ES. A subset of expression-site-associated genes (ESAGs) serves as signal for ES attenuation. Their depletion causes cell dormancy, metabolic silencing and gain of full developmental competence. The surprising connection between antigenic variation and developmental progression provides an unexpected point of attack against the deadly sleeping sickness.

11:30 AM - 11:45 AM (15 mins)
The trypanosome bilobe: form, fabric, and function
Brooke Morriswood[1], Katharina Havlicek[1], Heather Esson[2], Sevil Yavuz[1], Marco Sealey-Cardona[1], Graham Warren[1]
[1.]Max F. Perutz Laboratories, University of Vienna, Medical University of Vienna, Doktor Bohr-Gasse 9, 1030, Vienna, Austria. [2.]Present address: Laboratory of Evolutionary Protistology, Czech Academy of Science, University of South Bohemia, Ceske Budejovice, Czech Republic

Trypanosomes have a highly elaborate cytoskeleton that plays an integral part in cell viability, both in vitro and within their insect or mammalian hosts. The bilobe is an cytoskeletal structure of unclear function, defined by the marker protein TbMORN1. A combination of fluorescence microscopy and electron microscopy was used to build a precise morphological model of the bilobe in *Trypanosoma brucei*. These data showed that TbMORN1 describes a fishhook shape, encircling the top of the flagellar pocket above the flagellar pocket collar, and overlapping with the posterior end of the flagellum attachment zone. Pioneering use was made of proximity-dependent biotinylation (BioID) to screen for TbMORN1 binding partners and near neighbours. This is a novel technique that allows for forward biochemical screens under native conditions, employing a biotin ligase (BirA*) as a tag to promiscuously biotinylate proteins in the vicinity of the tagged candidate in a proximity-dependent fashion. Thus, the pattern of biotinylation preserves spatial information independently of lysis conditions or interaction strength. A total of 9 new proteins were found to colocalise, or partially overlap, with TbMORN1, representing a true positive rate of 90% in candidate evaluation. Depletion of TbMORN1 in procyclic and bloodstream form *T. brucei* cells showed that it is required for bilobe integrity. Loss of TbMORN1 is rapidly lethal in bloodstream form cells, demonstrating an essential role in vitro. Combining these phenotypic data with those relating to morphology (form) and composition (fabric), a possible model implicating the bilobe in the regulation of flagellar motility will be proposed.

11:45 AM - 12:00 PM (15 mins)
The trans-membrane protein homologue GPR89 promotes the development of stumpy forms in *Trypanosoma brucei*
Federico Rojas, Rachel Milne, Joanne Thompson, Keith R Matthews,
University of Edinburgh

G protein-coupled receptors are seven transmembrane domain (TM) proteins that transduce signals through their interactions with extracellular ligands and G protein-dependent and -independent signaling cascades allowing cells to respond to changes within their environment. Although trypanosomes are conventionally described as lacking GPCRs, we have identified a *T. brucei* protein (TbGPCR-like 1; TbGPCR-L1) related to the GPR89 family members that have been implicated in abscisic acid signaling in plants and Golgi acidification in mammals. The *T.brucei* protein has eight predicted

TM and localizes to the cell plasma membrane. Western blot and immunofluorescence analysis shows that in pleomorphic trypanosomes (i.e. capable of generating slender and stumpy forms), TbGPCR-L1 is expressed on slender forms but levels decrease as cells differentiate into intermediate and stumpy forms. In pleomorphic parasites, in contrast to monomorphic cells, inducible overexpression of TbGPCR-L1 protein drives premature differentiation in vitro and in vivo, generating stumpy cells at lower cell density compared to uninduced or the parental cell line. Overexpression of TbGPCR-L1 in a RNAi background targeting RBP7- a putative RNA binding protein required for normal quorum sensing- cells do not differentiate into stumpy cells, suggesting that TbGPR89 signalling acts upstream of RBP7 but on the same pathway. TbGPR89-L1 is rapidly degraded when overepxressed in what it seems to be an ubiquitin-dependent process involving the C-terminal portion of the protein, similar to the desensitization of GPCRs in other organisms. Our experiments indicate that TbGPR89-L1 might be expressed on slender forms as a sensor of an external stimulus thus initiating stumpy

12:00 PM - 12:15 PM (15 mins)

VSG identity and structural integrity determine growth rate of bloodstream form *Trypanosoma brucei*

Angela Schwede, Mark Carrington
University of Cambridge Department of Biochemistry Building, Downing Site Cambridge CB2 1QW UK

Trypanosome antigenic variation is based on extreme sequence variation in VSGs. How does the trypanosome accommodate different rates of VSG synthesis, folding and maturation? One possibility is that the VSG is always synthesised in excess so that growth is never limited by the identity of the VSG. Alternatively, the trypanosome could adapt growth rate to match the rate of VSG synthesis or VSG coat integrity. Here, we show that growth rate is determined by the identity of the VSG. The growth rate of a cell line expressing a 'slow' VSG was increased by ectopic expression of a 'fast' VSG transgene. In contrast, growth was dramatically slowed by the ectopic expression of VSG transgenes containing structural mutants. This phenotype was stable for several weeks without loss of viability and was fully reversible, reversion to normal growth resumed on inactivation of the transgene. Further, growth rate could be altered within hours using tetracycline inducible VSG transgenes. These results show that the trypanosome can adapt its growth rate in response to the VSG. A comparison of slow and fast growth transcriptomes was used to identify possible effectors of growth rate. Three of the findings are: (i) the slow growth transcriptome is different to the 'stumpy' transcriptome. (ii) One possible effector, PIP39, was validated and expression in bloodstream forms is inversely proportional to growth rate. (iii) Amongst the 47 most regulated mRNAs, 14 are involved in glucose metabolism. The upregulation of glycolysis in fast growing trypanosomes resembles the Warburg effect observed in cancer cells.

12:15 PM - 12:30 PM (15 mins)

Uncoupling flagellum formation and maintenance *

Cecile Fort, Philippe Bastin
Trypanosome Cell Biology Unit - CNRS URA 2581 Parasitology & Mycology Department Institut Pasteur, 25, rue du Docteur Roux, 75015 Paris

The length of the flagellum in *Trypanosoma brucei* varies from 3 to 30 µm according to the stage of its life cycle. It has been proposed that length is controlled by means of intraflagellar transport or IFT (Marshall et al. J. Cell Biol. , 2001). This is the transport of protein complexes or trains driven by kinesin motors from the base of the flagellum to the distal end (anterograde transport) that delivers tubulin for elongation of the organelle. The IFT trains come back to the base of the flagellum via retrograde transport. Knocking down the expression of any IFT protein inhibits flagellum formation. Intriguingly, flagella of intermediate length are still assembled at early time points of the RNAi induction. What happens in these short flagella during this period? To answer this question, we transformed the IFT88RNAi strain with a construct allowing endogenous tagging of IFT81, another IFT protein, with YFP. In non-induced conditions, robust IFT is observed in all flagella. When cells assemble shorter flagella one day after the induction of IFT88RNAi, IFT trains are less frequent and appear smaller. Moreover, IFT is not detected in old flagella that nevertheless maintain a normal length, meaning that IFT would not be necessary for their maintenance. We propose that the number of available IFT trains at the time when flagellum construction is initiated controls the length of the organelle. This is currently investigated by analysis of different IFT mutants and by photoconversion experiments.

Session B4 (WRR) - Apicomplexa - Translation

Chair: Julian Rayner, Wellcome Trust Sanger Institute

11:00 AM - 11:30 AM (30 mins)

Towards the Development of a Broadly-Neutralising Vaccine against Blood-Stage *Plasmodium falciparum*

Simon Draper

Jenner Institute, University of Oxford, UK

Recent results from the Phase III clinical trial of the leading pre-erythrocytic malaria vaccine candidate RTS,S, confirm that there remains a pressing need for the development of new and complementary approaches to vaccinate against *P. falciparum*. Vaccines against the parasite's asexual blood-stage have the potential to reduce mortality, morbidity and transmission of malaria, however no vaccine using the blood-stage antigens apical membrane antigen 1 (AMA1) or merozoite surface protein 1 (MSP1) has proven convincingly protective in clinical trials. Significant challenges have included antigenic polymorphism, the apparent requirement for exceptionally high antibody concentrations to mediate protection, and clinical-grade production of conformationally-accurate recombinant protein antigens. We have previously reported that vaccines based upon the full-length reticulocyte-binding protein homologue 5 (RH5) induce antibodies which neutralise all tested laboratory-adapted parasite lines, and have shown that neutralisation of recently-isolated parasites by anti-RH5 antibodies is more potent than with anti-AMA1 antibodies. We have also confirmed that vaccines based on RH5 can mediate robust heterologous strain efficacy in a stringent *Aotus nancymaae* non-human primate – *P. falciparum* challenge model. These data support the prompt clinical testing of RH5-based vaccines. This talk will describe the on-going development of new RH5 vaccine candidates that are progressing towards Phase I/IIa clinical testing.

11:30 AM - 11:45 AM (15 mins)

Development of Transgenic Rodent Malaria Parasites for Assessment of Novel Liver-Stage Malaria Vaccines *

Ahmed M. Salman[1,2], Rhea J Longley[1], Alexandra J. Spencer[1], Shahid M. Khan[2], Chris J. Janse[2], Adrian V.S. Hill[1]

[1.]*The Jenner Institute, University of Oxford, United Kingdom;* [2.]*Leiden Malaria Research Group (LMRG), LUMC, Leiden, The Netherlands*

At present there is no completely effective/licensed malaria vaccine. Most of the severe malaria pathologies/deaths are associated with *P. falciparum* strains, and developing effective vaccine remains a priority. Unfortunately, Pf doesn't infect small animals and some of the Pf candidate vaccines/antigens either differ from or are absent in rodent-parasites, limiting pre-clinical efficacy studies in murine-models. In this work we sought to rank/order the protective immune responses to several novel Pf vaccine-candidates using a novel rodent challenge-model. We first immunized mice with different Pf pre-erythrocytic vaccine-antigens delivered using viral vectored vaccines (ChAd63/MVA prime-boost) approach and tested the responses to these antigens in mice using this novel challenge-model. Specifically, we created transgenic *P. berghei* parasites expressing the Pf vaccine-candidate genes of interest thus enabling a vaccine efficacy/challenge assessment, *in vivo*. Consequently, we performed a screening of eleven Pf vaccine-candidates and rank/ordered them based on protection studies in different mice strains, selecting the most promising ones to be taken to further clinical trials. We created two mutants' sets: (i)Pf candidate-antigen genes were expressed in sporozoite and liver-stage using the Pbuis4 promoter; and (ii) when the Pf antigen had a Pb homolog, the Pb gene was replaced by its Pf equivalent and expressed under the corresponding Pb promoter. Both mutants' sets were used in the immunization/challenge studies. Based on results from our initial candidate-antigens immunogenicity 'ranking' experiments we have also created 'double-transgenic' parasites that express different combinations of the most promising candidates. These reagents are powerful tools for rapid assessment of multiple-antigen vaccines combinations.

11:45 AM - 12:00 PM (15 mins)

Efficacy of dihydroartemisinin-piperaquine in asymptomatic malaria parasite carriers: an evaluation of molecular markers of drug resistance

Mary C. Oguike, Bismarck Dinko, Colin J. Sutherland

Immunology and Infection Department, London School of Hygiene and Tropical Medicine; University of Health and Allied Sciences, Ghana

Asymptomatic malaria infection is a common phenomenon in areas of stable transmission. These infections can occur at microscopically detectable levels but mostly below threshold limit, thus, they serve as a continuous reservoir for malaria transmission. Artemisinin combination therapy is the current WHO recommendation in many countries but most clinical trials in endemic regions target *P. falciparum* paying less attention to *P. malariae* and *P. ovale* spp. ACTs are known to be highly efficacious against all *Plasmodium* species. In a recent study of *P. falciparum* gametocyte carriage and antibody responses, asymptomatic Ghanaian school children (274) were enrolled into a longitudinal study. Adequate dose of DP was given to children (152) with microscopically confirmed *P. falciparum* and followed up for 21 days. As a subsidiary analysis, molecular genotyping was done on the filter paper blood spots collected from this study. Interestingly, 215 of the 270 evaluable samples at enrolment tested positive for at least one specie (Pf, Pm, Poc or Pow). At day 21, twenty-two samples still tested positive (16Pf alone, 6 mixed species). Using bioinformatic tools, we designed new crt primers for *P. ovale curtisi, P. ovale wallikeri* and *P. malariae*. We then assessed polymorphisms in mdr1 and crt genes in these samples to ascertain the genotype of the pre- and post-treatment parasites. Results will be presented here.

12:00 PM - 12:15 PM (15 mins)

Emetine dihydrochloride hydrate: a potential candidate for repositioning in malaria *

Holly Matthews[1], Maryam Idris-Usman[1], Martin Read[2], Farid Khan[3] and Niroshini Nirmalan[1]

[1.]School of Environment and Life Science, University of Salford, Manchester, M5 4WT, United Kingdom; [2.]Faculty of Life Sciences, Manchester Interdisciplinary Biocentre, 131 Princess Street, Manchester, M1 7D, United Kingdom; [3.]Protein Technologies Ltd (PTL). Williams House, Manchester Science Park, Lloyd Street, North Manchester, M15 6SE, United Kingdom.

Drug repositioning, or the screening of existing drugs for new diseases, is increasingly viewed as an alternate strategy to fast-track drug discovery. Traditional drug discovery pipelines report ever increasing attrition rates despite the costly and time consuming processes involved. The vast repertoire of potential repositioning candidates with proven bioavailability, safety profiles and comprehensive historical information, in the public domain, will prove a vital resource to formulate mechanistic hypotheses on putative targets. Phenotypic screening, using the versatile SYBR Green-based flow cytometric and microtitre assays, of ~700 compounds, selected from two patent-expired drug libraries, yielded a complement of potential antimalarial leads (~ n=60) displaying strong in vitro inhibition of the *P. falciparum* K1 strain. Screening data highlighted the anti-amoebic drug emetine dihydrochloride as a potent antimalarial option. The work corroborates a previous study (Lucumi *et al.,* 2010) where IC50 values of 1 nM were reported in the drug sensitive 3D7 strain of *P. falciparum*. Our data on the multidrug resistant strain reports the retention of nanomolar efficacy with IC50 values of 47 nM. Despite concerns about toxicity, the nanomolar antimalarial potency, chemical malleability, the killing rate and combinatorial potential of the compound validates its further investigation as a standalone or combinatorial antimalarial candidate. A synthetic modification of the drug (dehydroemetine (Roche)) which structurally differs from emetine dihydrochloride only in a double bond next to the ethyl substituent is reported to retain its anti-amoebicidal properties while producing fewer side effects. Data from fixed-dose and CalcuSyn-based synergy studies and PRR/PCT experiments will be presented.

Session C4 (WRL) - Evolution I

Chair: Mark Field, University of Dundee

11:00 AM - 11:30 AM (30 mins)

Diversity and phylogeny of insect trypanosomatids

Julius Lukeš[1,2,*], Jan Votýpka[1,3], Tomáš Skalický[1,2], Jiří Týč[1,2], Dmitri A. Maslov [4] & Vyacheslav Yurchenko[1,5]

[1.]Biology Centre, Institute of Parasitology, Czech Academy of Sciences, and [2.]Faculty of Science, University of South Bohemia, České Budějovice (Budweis), Czech Republic; [3.]Faculty of Sciences, Charles University, Prague, Czech Republic; [4.]Department of Biology, University of California, Riverside, USA; [5.]Faculty of Science, Life Science Research Centre, University of Ostrava, Ostrava, Czech Republic

Monoxenous (= single-host) trypanosomatids represent a poorly studied, yet potentially very diverse and species-rich group within the family Trypanosomatidae. We have used several nuclear-encoded genes to reconstruct phylogenetic relationships among dozens of monoxenous trypanosomatids isolated from dipteran, heteropteran and siphonapteran insects captured at all continents except Antarctica. Some flagellates are confined to a single host with a cosmopolitan distribution, while others switch easily among host species, yet are confined to a small territory. Moreover, we have identified new clades, such as the flea-associated genus *Blechomonas*. The current inclusion of *Paratrypanosoma* that branches between the free-living bodonids and obligatory parasitic trypanosomatids shows that all three dixenous (= two-host genera (*Trypanosoma, Leishmania* and *Phytomonas*) are derived from closely related monoxenous lineages. Therefore, analysis of the genomes of the latter flagellates should provide insight into the emergence of the extremely successful life style of medically and economically important trypanosomatids.

11:30 AM - 11:45 AM (15 mins)

Evolution of chloroplast RNA processing at the boundary between photosynthetic and parasitic apicomplexans *

Richard Dorrell, R. Ellen R. Nisbet, James Drew, and Christopher J. Howe

RGD RERN JD CJH: Department of Biochemistry, University of Cambridge RERN: School of Pharmacy and Medical Sciences, University of South Australia

Chloroplast genome evolution is characterised by gene loss. Some of the most dramatic examples of this process are found within lineages that contain chloroplasts, but have lost the capacity to support themselves through photosynthesis, and instead often engage in parasitic life strategies. For example, apicomplexan parasites, such as the malaria pathogen *Plasmodium*, possess a secondarily non-photosynthetic chloroplast derived from a red algal endosymbiont, with a genome that only contains genes of housekeeping function, and no longer retains photosynthesis genes. Understanding the factors that contributed to this profound bifurcation in chloroplast genome content might provide insights into the evolution of such different lifestyle strategies in each lineage. Recently, two fully photosynthetic apicomplexans have been identified, the 'chromerid' algae *Chromera velia* and *Vitrella brassicaformis*, which retain photosynthesis genes within their chloroplasts. We have characterised chloroplast transcript processing pathways in both chromerid species, and in *Plasmodium falciparum*. We demonstrate that a transcript processing pathway, 3' poly(U) tail addition, is widespread across chromerid chloroplasts, and is principally associated with genes that function in photosynthesis, over transcripts of housekeeping genes. To our knowledge, this represents the first chloroplast transcript processing pathway to be associated with a particular functional category of genes. Poly(U) tail addition in chromerids is associated with high levels of transcript abundance, and plays an important role in directing transcript cleavage events on polycistronic precursor transcripts. In contrast, *Plasmodium* chloroplast transcripts are not polyuridylylated. Changes to the chloroplast transcript processing machinery may have underpinned the loss of photosynthesis in ancestors of parasitic

11:45 AM - 12:00 PM (15 mins)

Applied evolution: an experimental approach investigating how drug dosage affects the rate of resistance evolution *

Alan Reynolds, Jan Lindstrom, Paul Johnson, and Barbara Mable.

University of Glasgow, Institute of Biodiversity, Animal Health and Comparative Medicine. Graham Kerr Building, Glasgow, G12 8QQ.

The problem of overcoming resistance involves finding methods of drug use such that parasite populations are kept at low numbers and the evolution of resistance is minimised. Several factors are known to affect the rate at which parasites can evolve resistance, including the type of drug, dosage, timing of application, migration rates between susceptible and resistant populations, the standing frequency of resistance alleles in the population, and the specific mechanisms of resistance. In addition, life history characteristics of parasites may interact with the rate at which resistance develops. Current research to date on parasitic organisms has considered many of these factors in isolation but there has been little attempt to explore interactions between life-history traits and other factors affecting the rate of resistance evolution. This study takes an experimental evolutionary approach to understanding the influence of such interactions, using free-living *Caenorhabditis remanei* as a model. The rate of resistance evolution was evaluated by monitoring survival of populations treated with anthelmintics, applied at different dosages. In addition, changes in associated life-history traits and their trade-offs were quantified to understand the potential costs incurred by evolving resistance, under the different treatment dosages.

12:00 PM - 12:15 PM (15 mins)

Expansion and sculpting of the membrane trafficking system in the neuropathogenic amoeba *Naegleria fowleri*

Emily Herman[1], Alexander L. Greninger[2], Francine Marciano-Cabral[4], Govinda S. Visvesvara[3], Charles Y. Chiu[2, 5, 6], and Joel B. Dacks[1]

[1.]Department of Cell Biology, Faculty of Medicine and Dentistry, University of Alberta, Edmonton, Alberta, Canada; [2.]UCSF-Abbott Viral Diagnostics and Discovery Center, University of California San Francisco, San Francisco, California; [3.]Division of Foodborne, Waterborne and Environmental Diseases, National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, Georgia; [4.]Department of Microbiology and Immunology, Virginia Commonwealth University School of Medicine, Richmond, Virginia; [5.]Department of Medicine & [6.]Department of Laboratory Medicine, Division of Infectious Diseases, University of California San Francisco, San Francisco, California

Naegleria fowleri is an opportunistic pathogen in the supergroup Excavata. It is found in stagnant tropical, subtropical, and thermal waters around the world. It causes primary amoebic meningoencephalitis, killing 99% of those infected, usually within two weeks. Infection occurs when contaminated water enters the nose (e.g. when swimming), and *N. fowleri* passes through the cribriform plate to the olfactory bulb in the brain. *N. fowleri* is the only species of *Naegleria* that regularly infects humans, and no clear pathogenicity factor has been identified. We have sequenced the *N. fowleri* genome, and are using comparative genomics to identify differences between it and that of its harmless relative, *Naegleria gruberi*, in order to understand *N. fowleri*'s unique pathogenic ability. Due to its possible role in host invasion, we have performed comparative genomics of the membrane trafficking system (MTS), a defining feature of eukaryotic cells that is responsible for moving protein cargo between various organelles, and both into and out of the cell. We have observed multiple paralogous expansions of MTS components when compared with the non-pathogenic *N. gruberi*. Furthermore, the degree of these expansions does not correlate strictly with one or more genome duplication events, suggesting significant sculpting of the MTS in *N. fowleri*, which may play a role in its pathogenesis. We will provide an update of our ongoing complete genome analysis of the deadly *N. fowleri*.

12:15 PM - 12:30 PM (15 mins)

Streamlined endocytosis in *Trypanosoma brucei*

Paul Manna, Catarina Gadelha, Guido Van-Mierlo, Mark C. Field
College of Life Sciences, University of Dundee

To aid in immune evasion, mammalian infective form *Trypanosoma brucei* uses rapid endocytic turnover of its surface coat to remove host antibodies. Underlying this phenomenon is the evolutionarily ancient and widely conserved clathrin mediated endocytosis (CME) pathway. This system has been well studied in a number of common model organisms revealing a cohort of important gene products and plasma membrane lipids. Genetic comparisons suggest a minimisation of the CME system in *T. brucei*, at least in terms of the diversity of molecular components. We have extended these earlier comparative analyses to identify putative *T. brucei* endocytic proteins. Functional analysis of these candidates supports a streamlined mechanism of endocytosis in *T. brucei*, increasing speed and capacity by dispensing with time consuming cargo selection and concentration. However, the fundamental organisation of the system remains largely conserved and an evolutionary analysis demonstrates how dramatic effects at the systems level have arisen via gradual sculpting of components.

Session D4 (RCA) - Helminths - Genomic, Proteomic & Glycomic Biology

Chair: Cornelis Hokke, University of Lieden

11:00 AM - 11:30 AM (30 mins)

How do 'omics' technologies help us to control nematode infections?

Peter Geldhof

Laboratory for Parasitology, Department of Virology, Parasitology and Immunology, Faculty of Veterinary Medicine, Ghent University, Belgium

Molecular type research has gained huge momentum over the last decade by the development and improvement of several methodologies for both DNA and RNA sequencing and protein and glycan analysis. The application of these technologies in our research on the bovine gastrointestinal parasites *Ostertagia ostertagi* and *Cooperia oncophora* and the porcine intestinal parasite *Ascaris suum* has significantly advanced our understanding of the biology of these parasites and their interaction with the host. As an example, transcriptomic and proteomic analysis of A. suum larvae during their hepato-tracheal migration indicated that the degradation of complex carbohydrates likely forms an essential part of the energy metabolism of this parasite once it establishes in the small intestine. More than 30 glycosyl hydrolase encoding sequences are present in the *A. suum* genome, many of which are highly expressed on the intestinal surface of adult worms. Current research is focussed on evaluating their potential as vaccine and/or drug targets. As for *O. ostertagi* and *C. oncophora*, proteomic and glycomic analyses have been used to determine the structural composition of the activation-associated secreted protein (ASPs)-based vaccines we have previously developed for these parasites. The analyses indicated that the native ASPs are folded in a complex matter through the formation of at least 5 intra-molecular disulphide bridges and that they carry several N-linked glycans. The importance of all these features in the induction of a protective immune response is currently further investigated.

11:30 AM - 11:45 AM (15 mins)

***Fasciola hepatica*: in vitro maintenance and evaluation of unique tegumental proteins as novel control targets ***

Paul McCusker[1], Hayley Toet[2], Aaron G. Maule[1], Paul McVeigh[1], Erin McCammick[1], Colin C. Fleming[1], Angela Mousley[1], Terry W. Spithill[2], Nikki J. Marks[1]

[1.] Institute for Global Food Security, School of Biological Sciences, Queen's University Belfast, UK; [2.] Department of Agricultural Sciences and Centre for AgriBioscience, La Trobe University, Bundoora, Australia

The liver fluke (*Fasciola* spp.) costs the global agri-food industry ~$3.2 billion every year and up to 17 million humans are infected. Resistance to triclabendazole (the flukicide of choice) is increasing, contributing to the need for novel drugs and / or vaccines. We have been investigating five Fasciola-specific tegument proteins (designated Fh-TEG-1 to Fh-TEG-5) that have targetappeal due to their potential for surface expression and their lack of homology to host proteins. Using qPCR these genes were monitored in newly excysted juveniles (NEJs) over the 3-week period post-excystment -only Fh-teg-3 and Fh-teg-5 were expressed immediately following excystment, although during subsequent in vitro maintenance, Fh-teg-1 and Fh-teg-5 showed rapid upregulation. Importantly, all five tegumental genes are expressed in adults; mirroring independent RNAseq data for both immature-stage and adult F. hepatica. RNA interference (RNAi) of Fh-teg-1 and Fh-teg-5 in NEJs across a variety of time points was used to ascertain their importance to worm biology in vitro. No aberrant survival phenotypes were detected during in vitro maintenance. Ongoing studies on Fh-Teg RNAi-worms are examining tegument structure using electron microscopy and generating recombinant Fh-TEG-1 and Fh-TEG-5 to facilitate vaccine trials. For many helminth parasites, the inability to maintain them in vitro for extended periods limits opportunities for experimental manipulation. Here we also report our efforts to maintain liver fluke *in vitro* to advance the exploitation of its susceptibility to RNAi-based interventions. So far, we are able to maintain *in vitro* juvenile worms in excess.

11:45 AM - 12:00 PM (15 mins)

***Schistosoma mansoni* methyl-CpG binding domain protein (SmMBD2/3): a functional component of the schistosome epigenetic machinery ***

Sabrina Munshi, Karl F. Hoffmann
IBERS, Aberystwyth University

The fundamental components of the eukaryotic epigenetic machinery are DNA methyltransferases (Dnmts) and methyl-CpG binding domain proteins (MBDs). Dnmts enzymatically convert cytosines into their methylated form, 5-methylcytosine (5mC). MBDs are commonly known to target and bind 5mC and recruit various co-repressor complexes to these methylated genomic loci, leading to localised chromatin remodeling and transcriptional repression. Together, these epigenetic components are responsible for gene expression control and heritable alterations in phenotypic diversity. Previous studies in our laboratory have identified transcriptionally co-regulated Dnmt and MBD homologues (SmDnmt2 and SmMBD2/3) in the pathogenic trematode *S. mansoni*. Subsequent investigations have shown that SmDnmt2 is indeed a functional cytosine methyltransferase in this organism. Here, using DNA binding assays, yeast 2-hybrid screens and immunolocalisation, we present a comprehensive investigation of SmMBD2/3 function that suggests this protein is also a component of the *S. mansoni* epigenetic machinery. Combined with prior SmDnmt2 data, our characterisation of SmMBD2/3 function suggests that S. mansoni uses a primitive mechanism of DNA methylation as an additional level of gene control throughout the parasite life cycle.

12:00 PM - 12:15 PM (15 mins)

Characterization of excretory/secretory products and small metabolites of the pig whipworm *Trichuris suis*: insights on immunomodulation by intestinal parasites

Louis-Philippe Leroux[1], R Valanparambil[2], PP Gros[1], M Mitreva[3], MM Stevenson[2], JF Urban[4], JV Weinstock[5], TG Geary[1], A Jardim[1]
[1.] Institute of Parasitology, McGill, Canada; [2.] McGill Centre for the Study of Host Resistance, McGill University Health Centre, Canada; [3.] Genome Institute, Washington University, USA; [4.] U.S. Department of Agriculture, USA; [5.] Tufts University, USA.

The incidence of allergic and auto-immune disorders in industrialized countries has increased dramatically over the past century. Epidemiologic studies have linked this trend to the reduction of infectious diseases and the modern hygienic lifestyle, an explanation dubbed the “hygiene hypothesis”. This assumption is validated in part by the growing evidence showing that helminths may play a protective role in the infected host by inducing a balance between the Th1 and Th2 branches of the immune system and preventing immunological disorders. The pig whipworm *Trichuris suis* has become an interesting model to study this phenomenon. Studies have clearly shown that *T. suis* suppresses clinical symptoms of several autoimmune diseases such as inflammatory bowel diseases (IBD), multiple sclerosis (MS), and possibly autism. *T. suis* excreted/secreted (ES) molecules have immunomodulatory properties and are likely implicated in these immunotherapeutic events. To identify the immunoactive components, we performed proteomic analyses on ES collected in vitro from different life stages of the parasite. Tandem-mass spectrometry (MS/MS) analyses generated a list of ~300 proteins with temporal expression profiles. Treating immune cells in vitro inhibited stimuli-induced production of pro-inflammatory cytokines, namely IL-12 and TNF-alpha. Interestingly, ES alone induced expression of the anti-inflammatory cytokine IL-10. Finally, solid phase extractions of parasite-conditioned culture media confirmed that small molecular weight metabolites released by the worms have immunomodulatory properties that could act in concert with proteins to modulate host immune functions. A better understanding of how helminths affect the hosts' immune system may help improve therapeutic strategies against autoimmune diseases.

12:15 PM - 12:30 PM (15 mins)

The whipworm genome and transcriptome

Bernardo Foth, Isheng J. Tsai, Adam J. Reid, Allison J. Bancroft, Sarah Nichol, Alan Tracey, Nancy Holroyd, James A. Cotton, Eleanor J. Stanley, Magdalena Zarowiecki, Thomas Huckvale, Philip J. Cooper, Richard K. Grencis, Matthew Berriman
Parasite Genomics, Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SA, UK. Division of Parasitology, Department of Infectious Disease, Faculty of Medicine, University of Miyazaki, Miyazaki 889-1692, Japan. Faculty of Life Sciences, University of Manchester, Oxford Road, Manchester M13 9PT, UK. Liverpool School of Tropical Medicine, Pembroke Place, Liverpool L3 5QA, UK. Centro de Investigación en Enfermedades Infecciosas, Escuela de Biología, Pontificia Universidad Católica del Ecuador, Quito, Ecuador.

Whipworms are common soil-transmitted helminths that cause debilitating chronic infections in man. These nematodes are only distantly related to *Caenorhabditis elegans* and have evolved to occupy an unusual biological niche, tunneling through epithelial cells of the large intestine. Here we present the genome sequences of the human-infective *Trichuris trichiura* and the murine laboratory model *T. muris*. We identify the sex chromosomes and centromeric repeats and find only limited synteny between the species. Whole transcriptome analyses in *T. muris* detect many genes and protein families that are expressed in a gender- or life stage-specific manner, such as WAP domain-containing or DNase II-like proteins. Together, they characterise the transcriptional landscape of a morphological region with unique biological adaptations, namely bacillary band and stichosome, found only in whipworms and related parasites. These genomes and associated functional data elucidate key aspects of these nematodes and their molecular host-parasite interactions and provide a solid foundation for future studies, facilitating the development of novel anthelmintic interventions and the exploitation of unique whipworm biology.

Session E4 (RCU) - Public understanding, outreach and communication - Presenting Science to the Public - On Their Terms

Chair: Tansy Hammarton, University of Glasgow

11:00 AM - 11:30 AM (30 mins)
Presenting science to the public - on their terms
Mike Barrett
Wellcome Trust Centre for Molecular Parasitology and Glasgow Polyomics, University of Glasgow. Glasgow G12

Many people still consider Science to be difficult, impenetrable, jargon-rich and of little importance to their every-day lives. Presenting scientific concepts to the public is best achieved by demonstrating the importance of science in ways that make its impact apparent whilst avoiding the terminology associated with the fears installed by its complexity. Relating scientific discoveries to topical subjects is a good way to make discoveries relevant, for example by linking celebrities or key events to scientific topics. Here I will give a light-hearted overview of ways to emphasise the importance of research into Parasitic disease to non-specialist audiences.

11:30 AM - 11:45 AM (15 mins)
Crafty Critters: crafting a parasite infestation *
Rebecca Devlin, Catarina A Marques, Robyn Kent, Nicola J Mamczur, Zara Gladman, Mhairi Stewart
University of Glasgow, 120 University Place, Glasgow G12 8TA

The Crafty Critters project will be creating an infestation of parasites and other microorganisms in fluffy form! Developed by students and staff in partnership with Glasgow Science Festival, Crafty Critters aims to spread the word on the fascinating parasites studied at Glasgow. We will be holding multiple crafting workshops in 2014, where people will be invited to crochet or felt-craft a fluffy parasite and chat with scientists about the parasites they're creating. The project will engage a range of audiences with science: adults, families and children, while also creating teaching aid materials. These materials can later be used in other public engagement events by schools and sent abroad to endemic countries. Crafty Critters will launch on 14th June (World Knitting Day), with a workshop during Glasgow Science Festival and more events to be announced, finishing with a 'parasitic infestation' display in October 2014. We will talk about the challenges in funding and organising a large series of events such as these and the skills developed as a result.

11:45 AM - 12:00 PM (15 mins)
Communicating science to the public across the generations
Linda Kohl
UMR7245, National Museum of Natural History, Paris, France

The National Museum of Natural History in Paris has a longstanding tradition of promoting Science and communicating with the general public. Almost 25 years ago the Museum initiated a first project on ‘Participating Science’, where ornithologists reported the presence of common birds in a particular region over a determined time period, according to a standardized protocol. In recent years activities have sometimes been tailored towards specific groups: families, pupils, teachers and other adults. The majority of themes treated deal with Biodiversity, in France and in the World. Some of the programs are limited to a particular site, such as the Children’s Gallery located within the Gallery of Evolution in Paris. Other projects are national: during the 2013 ‘Tour de France” cycling race, each stage was used to highlight a specific plant or animal species of the region. During the Papoua-New Guinea expedition in 2013, over one hundred school classes took part in the adventure via Internet and learned about the work of researchers and biodiversity. Teachers can also book workshops and get up to date on topics such as biodiversity, classification and scientific techniques. Throughout the year conferences and discussions on topical subjects are held. During the yearly Science fair, the public is invited to participate in various demonstrations and exhibits taking place on the campus. Our workshop on unicellular organisms gives the visitors an overview ranging from cell structure, to diversity of unicellular organisms (free living and parasites), to the isolation of molecules produced by these organisms.

12:00 PM - 12:30 PM (30 mins)

Tapeworm Diaries

Philip Craig

Cestode Zoonoses Research Group, School of Environment and Life Sciences, University of Salford, Greater Manchester M5 4WT, UK.

Tapeworms have intrigued and fascinated us since antiquity due to their great size, our natural abhorrence of them, and the associated symptoms of spontaneous segment or strobila release. Not surprisingly only a handful of human voluntary self-infections have been reported in the literature from the 19th and 20th Centuries. However, the advent of science documentaries and reality-TV has resulted in the occasional focus on parasites and human parasitic infections. Such programmes have included tapeworm self-infection stories at least 4 times on UK television in the last 15 years (BBC2, BBC4, Channel 4, Sky). I will recount my own experience of self-infection with *Taenia saginata*, and my subsequent role as informal advisor to two TV presenters who also infected themselves with the beef tapeworm for a TV programme.

Session F4 (RCJ) - Antihelmintic overuse - evolution under pressure

Chair: Jacqui Matthews, Moredun Research Institute

11:00 AM - 11:30 AM (30 mins)
Title not received.
Andrew Read
Penn State

Abstract not received.

11:30 AM - 11:45 AM (15 mins)
The emergence of anthelmintic resistance in parasitic nematodes of livestock is characterised by multiple hard and soft selective sweeps of independently derived mutations
Libby Redman[1], Fiona Whitelaw[2], Andrew Tait[2], Charlotte Burgess[3], Yvonne Bartley[3], Philip Skuce[3], Frank Jackson[3], John Stuart Gilleard[1]
[1.] Department of Comparative Biology and Experimental Medicine, Faculty of Veterinary Medicine, University of Calgary, Calgary, Alberta, T2N 4N1 Canada. [2.] Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow G61 1QH, United Kingdom. [3.] Moredun Research Institute, Pentlands Science Park, Midlothian EH26 0PZ, United Kingdom.

Although anthelmintic resistance has been a major problem in livestock parasites for decades, our understanding of how a resistance mutations arise and spread in parasitic nematode populations is still limited. There is also little information on the genomic changes that occur as anthelmintic resistance mutations increase in frequency in parasite populations; the so-called “genetic signature” of selection. Such knowledge is critical if we are to be able to apply genome-wide population genomic approaches to identify anthelmintic resistance mutations. To address these questions, we investigated the population genetics of benzimidazole resistance of *T. circumcincta* and *H. contortus* on sheep farms in the UK. These two species were sampled from the same animals on the same farms allowing a direct comparison between the two species without any confounding differences in environment or drug treatment history. We have found an extremely high level of genetic diversity of resistance alleles in both parasite species. This diversity creates a soft selective sweep overall for both parasites but one which consists of a complex mosaic of harder and softer sweeps on individual farms. In spite of the complex pattern of selective sweeps, the genetic signature of selection can be detected using simple measures of genetic diversity and departures from neutrality. This has important implications for the application of genome-wide population genomic approaches to identify importance anthelmintic resistance conferring loci. We propose a model of parallel adaptation where independent mutations repeatedly arise on different farms by with subsequent mixing different resistance haplotypes by animal movement between farms.

11:45 AM - 12:00 PM (15 mins)
Help: the wormers don't work!
Jacqueline Matthews
Moredun Research Institute, Edinburgh EH26 0BL.

Parasitic helminths are ubiquitous in horse populations worldwide. In horses that graze contaminated pasture and which are not treated with appropriate anthelmintics, large numbers of can accumulate. These include immature larval stages that develop in the wall of the large intestine. A build up of larvae can lead to serious disease and death. Horse nematodes have been controlled using anthelmintics, of which three classes are available for use. In the developed world, resistance to two of these classes is widespread in some species and resistance is emerging to the third class, the macrocyclic lactones. Should multi-class resistance spread throughout the general equine population, there will be no options left for control, as no new anthelmintic classes are under development for horses in the short-medium term. It is therefore essential that rational deworming programmes, that aim to preserve efficacy of the remaining effective anthelmintics be instituted. This presentation focuses on recent knowledge garnered on targeted anthelmintic treatment strategies for equine helminth control and ways in which these protocols can be supported by appropriate diagnostics.

12:00 PM - 12:15 AM (15 mins)

Metabolic enzymes of the liver fluke, *Fasciola hepatica*: biochemical characterisation and identification of inhibitors

David Timson[1], Veronika L Zinsser[1], Steffen Lindert[2], Samantha Banford[1], Elizabeth M Hoey[1], Alan Trudgett[1]

[1.] School of Biological Sciences, Queen's University Belfast, Medical Biology Centre, 97 Lisburn Road, Belfast BT9 7BL. UK.

[2.] Department of Pharmacology and Center for Theoretical Biological Physics, University of California San Diego, La Jolla, CA, 92093. USA.

In theory, metabolic enzymes represent excellent targets for new drugs. Inhibition will result in reduced energy production and/or reduced biosynthesis both of which would be highly detrimental to the parasite. However, the high levels of sequence and structural similarity between parasite and host enzymes can present a barrier to the development of selective inhibitors. Nevertheless, a number of drug-like molecules which target metabolic processes have been identified in unicellular parasites. Thus, we are undertaking biochemical investigations on liver fluke enzymes in order to learn more about whether and how they differ from the equivalent mammalian proteins. We have shown that *Fasciola hepatica* triose phosphate isomerase (FhTPI) is a highly stable protein and its enzymatic activity is weakly inhibited by phosphoenolpyruvate. This stability is greater than the mammalian enzyme and the degree of inhibition is less. This suggests that there are subtle differences in structure and biochemistry which could be exploited in future drug discovery. *F. hepatica* UDP-galactose 4'-epimerase (FhGALE) is a dimeric enzyme with binds an NAD cofactor at each of its active sites. Its catalytic parameters differ from the human enzyme, especially the Michaelis constant for UDP-galactose which is ~5-fold higher. The structure of the enzyme has been modelled and this model used to identify potential inhibitors. The top 40 hits have been tested, and six shown to inhibit, two with considerable selectivity over the human enzyme. These compounds should be investigated further as leads for potential novel therapeutics.

12:15 PM - 12:30 PM (15 mins)

Pharmacokinetic/Pharmacodynamic modelling of anti-Wolbachia agents. *

David Waterhouse, Ghaith Aljayyoussi, Joseph Turner, Mark Taylor, Stephen Ward

Parasitology Department Liverpool School of Tropical Medicine Pembroke Place Liverpool Merseyside L3 5QA United Kingdom

Doxycycline is currently the gold standard for the targeting of Wolbachia in lymphatic filariasis and onchocerciasis chemotherapy. However, the current drug regimen is a 100-200 mg/day doxycycline dose given for 4 to 6 weeks to patients. The AWOL consortium funded by the Bill & Melinda Gates foundation aim to reduce the current treatment time to 7 days or less, to be compatible with mass drug administration programmes. To achieve a rapid 7-day or less kill rate of Wolbachia, a number of registered drug combinations will be employed. These include different tetracyclines (doxycycline and minocycline) rifamycins (Rifampicin or Rifapentine), moxifloxacin as well as anti-helminthic drugs. The complexity of multiple drug combinations necessitates a rational approach in the identification and choice of the best treatments in in-vivo models and translating the animal treatments in the lab into clinical trials in the field. We have performed a series of PK-PD models and simulations using multiple software programs such as Pmetrics®(Population non-compartmental analysis) and Stella® to further dissect and quantify the dynamics of anti-bacterial activity of these drugs in the treatment of lymphatic filariasis and onchocerciasis. As an example here, we identified the PK parameters of doxycycline, minocycline and rifampicin in in-vivo PK studies in the SCID mouse *Brugia malayi* model and used the PK data with pharmacodynamic outputs to interpret the PK-PD relationships in light of the effect of each drug upon Wolbachia viability in parasites. The data display's the power of PK-PD modelling in quantifying PK-PD relationships for testing in clinical trials.

Session A5 (WRA) - Kinetoplastida - Growth, form and metabolism II

Chair: Mike Barrett, University of Glasgow

2:00 PM - 2:30 PM (30 mins)

Comparison of the central metabolism of the insect and bloodstream trypanosomes

Frédéric Bringaud, Muriel Mazet, Yoann Millerioux

Centre de Résonance Magnétique des Systèmes Biologiques, UMR-5536 Université Bordeaux Segalen, CNRS, 146 rue Léo Saignat, 33076 Bordeaux, France

We have compared metabolism of two main carbon sources, glucose and threonine, used by the insect procyclic form (PF) and mammalian long-slender bloodstream form (LS-BSF) of *T. brucei* to produce their ATP and intermediary metabolites. PF primarily metabolizes glucose into the excreted acetate and succinate end products, while threonine is converted into acetate and glycine. Mitochondrial production of acetate from both carbon sources is synthetically essential for fatty acid biosynthesis and ATP production. LS-BSF developed a high glycolytic flux, ~10-fold higher compared to PF, with pyruvate being considered the only end product excreted from glucose metabolism. In contrast to the current view, we showed that LS-BSF produces almost as much acetate and succinate from glucose as PF. Acetate is also produced from threonine although at a lower rate compared to glucose-derived acetate. More importantly, acetate production from glucose and threonine is synthetically essential for growth of LS-BSF and abolition of succinate production from glucose is lethal for the parasites. These data highlight that the central metabolism of LS-BSF contains unexpected essential pathways, although minor in terms of metabolic flux compared to the glycolytic rate, which could be targeted for the development of trypanocidal drugs.

2:30 PM - 2:45 PM (15 mins)

Iron uptake in *Trypanosoma brucei*

Martin Taylor, A. McLatchie, D. Ntais, J.A. Thomas, J. M. Kelly

Department of Pathogen Molecular Biology, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine

Iron is an essential nutrient in trypanosomes. Bloodstream trypanosomes derive iron from host transferrin via the ESAG6/7 transferrin receptor. Iron is subsequently released from transferrin in the lysosome as FeIII. However, the pathway by which FeIII is reduced then transported into the cytosol has not been resolved. The minimal requirement for this process would be a ferric reductase and a cation channel/transporter. We have constructed bloodstream form null mutants of the two transmembrane ferric reductases (FRs) in the *T. brucei* genome. The FRs are orthologues of cytochrome b561 (TbCytb561) and cytochrome b558 (TbFre1), respectively. Both null mutants show no growth defect *in vitro*, however the TbCytb561 mutants (and TbCytb561 RNAi lines) are resistant to the iron chelator deferoxamine (DFO), while the TbFre mutants are more susceptible to DFO than the wild type. This apparent paradox is explainable by differential protein localization. We have also constructed cell lines in which both TbCytb561 and TbFre1 have been deleted following four rounds of gene deletion. These double mutants grow in vitro. The phenotype of these cells will be discussed.

2:45 PM - 3:00 PM (15 mins)

Variations in Swimming Patterns and Behaviour of African Trypanosomes in Mammalian Hosts Depicts Adaptation to Survive in Diverse Environments *

Joel Bargul[1, 2,] Francis A. McOdimba[3], Jamin Jung[4], Vincent O. Adung'a[1, 5], Timothy Krüger[4], Daniel K. Masiga[1], Markus Engstler[4]

[1.]Molecular Biology and Bioinformatics Unit, International Centre of Insect Physiology and Ecology, P.O. Box 30772, Nairobi, Kenya; [2.]Department of Biochemistry, Jomo Kenyatta University of Agriculture and Technology, P.O. Box 62000 Nairobi, Kenya; [3.]Modern Medical Diagnostic Consultants, P.O. Box 15172 Nairobi, Kenya; [4.]Department of Cell and Developmental Biology, Biocenter, University of Würzburg, Würzburg, Germany.

African trypanosomes are able to survive in harsh conditions in the vertebrate host, despite constant attacks by host

antibodies. They evade immune response through antigenic variation and incessant motility. Their motility patterns are highly variable in different environments. Findings of this study show that variations in swimming patterns, speeds and behaviour of these parasites represent adaptation to survival in different mammalian hosts. High-speed video microscopy was used to quantify motility in *Trypanosoma congolense*, rodent-adapted *T. vivax, T. brucei brucei* and *T. evansi*. The average swimming speed of *T. congolense* was higher in mouse than sheep blood, unlike *T. vivax* whose average speeds were similar in both hosts (p=0.5874). Mean speed reduction in *T. congolense* (in sheep wet blood films) was ascribed to adherence of most parasites to the erythrocytes. Both *T. vivax* (9%) and *T. congolense* (more than 85%) lacked adherence in murine blood. These variations in motility behaviour exhibited by the same parasite isolate in different vertebrate hosts show adaptation to survival. We hypothesize that the differences in swimming speeds influences antibody clearance. Better understanding of the trypanosome motility could lead to new ways of fighting these diseases, for example by disrupting flagellar-driven motility.

3:00 PM - 3:15 PM (15 mins)
Developing a flagellar transition zone proteome in *Trypanosoma brucei*
Samuel Dean, Keith Gull
University of Oxford, Sir Wiliam Dunn School of Pathology South Parks Road, OX1 3RE

The biology of the trypanosome flagellum provides opportunities for insights into trypanosome pathology and can be generalised to other eukaryotic flagella. The flagellum transition zone, positioned between the distal end of the basal bodies and the proximal end of the 9+2 axoneme, has increasingly become recognised as of central importance to flagellum growth and function. The transition zone and associated appendages act as a ciliary gate keeper, maintaining the different composition of the flagellum and cell body that is required for development and signaling. Many of the human inherited ciliopathy diseases, such as Meckel and Joubert syndrome, are now recognised as transition zone diseases. Despite this, few proteins have been identified that localise to the transition zone in trypanosomes, and the proteins that make up the characteristic structures of the transition zone (such as the Y shaped linkers that link the axoneme to the cell membrane, and transitional fibres that act as docking sites for IFT vesicles) are largely uncharacterised in any organism. I have used a novel strategy to purify the cytoskeletal architecture encompassing the transition zone. To date, I have identified >30 proteins that localize the transition zone and associated structures. These include novel components of the Y-shaped linkers, transitional fibers, flagellar pocket neck and bilobe structures as well as the B9 and BBsome ciliopathy complexes. Interestingly, however, many of the transition zone proteins identified in this study have no identifiable orthologs outside of kinetoplastids, highlighting the complexities of the evolutionary cell biology of this organelle.

3:15 PM - 3:30 PM (15 mins)
The surface landscape of African trypanosomes
Catarina Gadelha, Wenzhu Zhang, Brian T. Chait, Mark C. Field
University of Nottingham, The Rockefeller University, University of Dundee.

To perform antigenic variation, the African trypanosome surface coat must be kept free of many essential invariant proteins. These are instead sequestered to the flagellar pocket, a specialised region of the surface membrane that is the sole site of endo/exocytosis. In previous work we and others defined morphological membrane domains and boundaries around the flagellar pocket, identified their association with the internal cytoskeleton, and described how nutrient macromolecules may gain access to the cell interior via a continuous channel linking the extracellular environment to the pocket lumen. However, there remains a paucity of data concerning the molecular composition of the flagellar pocket. This severely hampers understanding of flagellar pocket mechanisms, and also possible exploitation in drug and vaccine development. Using chemical derivatisation of surface membrane, tandem mass spectrometry, quantitative analysis and bioinformatic filters we describe a new surface proteome for bloodstream form *Trypanosoma brucei*. This set is enriched in GPI-anchored proteins, trans-membrane proteins and annotations similar to known surface components. By creating a genetic toolkit for tagging membrane proteins from endogenous loci, we have localised several putative surface molecules of unknown function. Our results validate the surface membrane location of many novel components, and also show that individual proteins can access different combinations of cell body, flagellar and flagellar pocket membranes. We propose that *T. brucei* exhibits distinct domains on its surface with restricted diffusion between them. This paradigm has important implications for the function of the trypanosome cell surface.

Session B5 (WRR) - Apicomplexa - Pathogenesis

Chair: Catherine Merrick, Keele University

2:00 PM - 2:30 PM (30 mins)

Conserved structural features required for interaction with endothelial protein C receptor in severe malaria

Matthew Higgins[1], Clinton Lau[1], Louise Turner[2], Jakob S. Jespersen[2], Ed Lowe[1], Bent Petersen[2], Thor G. Theander[2], Thomas Lavstsen[2]

[1.]Department of Biochemistry, University of Oxford, South Parks Road, Oxford, OX1 3QU; [2.]Centre for Medical Parasitology, University of Copenhagen, Copenhagen, Denmark.

The PfEMP1 proteins of *Plasmodium falciparum* cause infected erythrocytes to adhere to endothelial surfaces and to tissues. This protects the parasite from spleen-mediated clearance and prolongs the infection. However, it also leads to specific pathology, with accumulation of infected erythrocytes within the brain causing cerebral malaria. As naturally acquired immunity to severe malaria correlates with the acquisition of antibodies that target the PfEMP1 proteins, it is important to understand which PfEMP1s cause severe disease and to develop vaccines that mimic this immunity. However, this is a great challenge, as the ~60 PfEMP1 protein genes in the parasite genome can bind to a variety of ligands and show little sequence identity. Last year, a correlation was demonstrated between development of severe malaria and the ability of infected erythrocytes to bind to endothelial protein C receptor (EPCR). This suggests a mechanism in which the binding of PfEMP1s blocks the binding the natural ligands of EPCR, preventing anti-inflammatory responses. Here we will present our studies of the PfEMP1-EPCR interaction and other PfEMP1 mediated binding processes, summarizing what we know about how PfEMP1s interact with their ligands.

2:30 PM - 2:45 PM (15 mins)

Alteration of the Blood-Brain Barrier (BBB) endothelial cells, secondary to *Plasmodium falciparum* infected red blood cells (PRBC) sequestration in Cerebral Malaria: an *in-vitro* study *

Mohd Hamzah Mohd Nasir[1], Monique F. Stins[2] and Srabasti J. Chakravorty[1]

[1.]Institute for Science and Technology in Medicine, School of Life Sciences, Keele University, Keele, Staffordshire, ST5 5BG, UK; [2.]John Hopkins University, Baltimore, USA

Cerebral Malaria (CM), a severe complication in *Plasmodium falciparum* infection is classified as one of the main causes of mortality in malaria. The clinical outcome of CM, broadly varies from recovery to a lethal endpoint. Investigation of post-mortem brain tissue of CM patients demonstrates BBB breakdown (microhaemorrhages) with loss of tight junction proteins (TJPs), occludin, claudin-5 and ZO-1 in endothelial cells. Interestingly, this was observed in vessels in the presence and in the absence of sequestered PRBC. This suggested the mobilisation of direct and indirect mechanisms following PRBC sequestration. In previous studies, the endothelial cell monolayer integrity was reduced when co-cultured directly with PRBC. To investigate the indirect effect of sequestration to the BBB integrity, human brain endothelial cells (tHBEC) were first co-cultured with PRBC after which, the co-culture supernatant was harvested and analysed for the presence of candidate soluble factors and functional assays performed. Interestingly, analysis of the co-culture supernatant showed the induction of the ADAMTS family of protease, ADAMTS-4. In addition, differential regulation of ADAMTS-1 and the matrix metalloproteases (MMP) family proteases, MMP-2 and MMP-9 was observed. Treatment of freshly cultured tHBEC monolayer with the co-culture supernatant demonstrated up to 2-fold reduction in tHBEC monolayer integrity within 3 hours, measured using FITC-dextran permeability assay. This was accompanied by reduction in the TEER and the endothelial cell TJPs. We propose that endothelial cell-derived proteases in the co-culture supernatants that are released as a result of interaction with PRBC, during sequestration; contribute to BBB breakdown in CM.

2:45 PM - 3:00 PM (15 mins)

Generation of antigenic diversity in *Plasmodium falciparum* by structured rearrangement of var genes

William L. Hamilton[1], Antoine Claessens[1], Mihir Kekre[1], Thomas D. Otto[1], Adnan Faizullabhoy[1], Julian C. Rayner[1], Dominic Kwiatkowski[2]

[1.]Malaria Programme, Wellcome Trust Sanger Institute, Hinxton, UK; [2.]Wellcome Trust Sanger Institute and MRC Centre for Genomics and Global Health, University of Oxford.

Hypervariable antigens encoded by the multi-copy var gene family are critical for pathogenesis and immune evasion in *Plasmodium falciparum*. We constructed and performed whole genome sequence analysis of large clone trees to study the generation of novel var gene sequences in asexually replicating parasites. 270 genomes were analysed from sub-clones that had been in culture for a combined total of >1,000 days, producing an order of magnitude more data than comparable previous studies. High rates of non-allelic recombination between different var genes, located both within chromosomes and at the subtelomeres, were detected. Recombination produced chimeric sequences through gene conversions and duplications, and was observed both in established lab strains and in recently culture-adapted Cambodian field isolates. Analysis of >100 recombination events involving var exon 1 revealed that the average nucleotide sequence identity of two recombining exons was only 63% (range: 52.7 - 72.4%) yet the crossovers were error-free and occurred in such a way that the resulting sequence was in frame and domain architecture was preserved. Var exon 1, which encodes the immunologically exposed part of the protein, recombined in up to 0.2% of infected erythrocytes in vitro per life cycle, indicating that millions of new antigenic structures could potentially be generated each day in a single infected individual, facilitating long-term parasite survival.

3:00 PM - 3:15 PM (15 mins)

A systems biology approach to characterizing host-parasite interactions during *Toxoplasma* cell invasion

Dong Xia, D.P. Beiting, M. A. Diaz-Miranda, Q. Zheng, D.S. Roos, A.R. Jones, B.D. Gregory, J.M. Wastling
University of Liverpool

Understanding host cell-pathogen interactions is crucial in determining the key factors controlling virulence and disease pathogenesis in *Toxoplasma gondii*. In this study we adopted a systems based approach to look simultaneously at dynamic modulations in both host and parasite transcriptomes and proteomes during *Toxoplasma* cell invasion. Our study was focussed on the early stages of cell invasion and used quantitative label-free proteomics and RNASeq to characterise protein and gene expression simultaneously from both host cells and parasites across various strains and life-cycle stages of *T. gondii*. This study permitted a parallel interrogation of host and pathogen responses, as well as a direct comparison of transcript and protein expression data. Results from clustering and network analysis identified dynamic changes in host and parasite responses during infection, as well as clusters of co-regulated proteins, enabling us to build a preliminary systems-model of host-parasite interactions. Statistical and functional bioinformatics analyses showed that transcriptional profiles did not always match quantitative proteomics data, which may indicate complex mechanisms of gene expression and regulation. The established workflow and data analysis pipeline are readily adaptable to study host-parasite interactions under various treatments or physiological conditions. The expression data acquired from both host and parasite have been integrated into EuPathDB and are now freely available to the user community.

3:15 PM - 3:30 PM (15 mins)

Strain-transcending antibodies against group A PfEMP1 variants implicated in severe childhood *Plasmodium falciparum* malaria

Ashfaq Ghumra, Ricardo Ataide, Ahmed Raza, J. Alexandra Rowe
Centre for Immunity, Infection and Evolution, Institute of Infection and Immunity, School of Biological Sciences, University of Edinburgh, Edinburgh, United Kingdom

Rosetting, the binding of infected erythrocytes (IE) to uninfected erythrocytes is a parasite adhesion phenotype associated with severe malaria. Parasite rosetting ligands on the IE surface include specific *P. falciparum* Erythrocyte Membrane Protein 1 (PfEMP1) variants encoded by group A var genes. We are studying *P. falciparum* parasites that show a virulence-associated rosetting phenotype. Although antibodies raised against the N-terminal PfEMP1 domain (NTS-DBLα) from Group A PfEMP1 variants have strain-transcending activity, it remains unknown whether other extracellular PfEMP1 domains are able to induce strain-transcending antibodies. In this study, antibodies were raised to all the extracellular PfEMP1 domains of the IT4var60 PfEMP1 variant, which is expressed by rosetting IT/PAR+ parasites. Rabbits were immunised with proteins expressed in E. coli. Antibodies to every domain were able to inhibit rosetting of the homologous IT/PAR+ parasite, although antibodies to the N-terminal NTS-DBLα domain were the most effective (IC50 0.08 μg/ml). The IT4var60 antibodies to all domains exhibited strain-transcending surface reactivity against live infected red cells of heterologous rosetting strains. Furthermore, the IT4var60 antibodies displayed functional cross-reactivity by rosette inhibition and phagocytosis of heterologous strains. Intriguingly, cross-reactivity was restricted to the rosette-mediating PfEMP1 variants that could also bind non-immune IgM. These data show that it is possible to raise strain-transcending antibodies against PfEMP1 variants that mediate the rosetting phenotype that is important in severe childhood malaria. Further work is to highlight shared epitopes between PfEMP1 variants and determine the potential for a vaccine against rosetting parasites to reduce the risk of severe malaria.

Session C5 (WRL) - Evolution II

Chair: Jamie Stevens, University of Exeter

2:00 PM - 2:30 PM (30 mins)
From endolysosomes to invasion organelles: evolutionary remodeling of membrane-trafficking across the Apicomplexa.
Joel Dacks
University of Alberta

The rhoptries and micronemes are secretory organelles crucial to the invasion process in apicomplexan parasites. Understanding the evolutionary path that has lead to these organelles both sheds light on the cellular evolution of parasitism and points the way towards a better molecular parasitological understanding of the invasion process. Functional and comparative genomic work support the proposed origin of rhoptries and micronemes as derived secretory endolysosomal organelles. This evolutionary shift has been accompanied by changes in the encoded complement of membrane-trafficking machinery in apicomplexan genomes, which we continue to explore. However, the apical complex predates the parasitic Apicomplexa, being found in dinoflagellates and the chromerids. Thus comparative genomic analysis allows us to begin teasing apart those modifications to the membrane-trafficking that are associated with the evolution of parasitism and those that may have been pre-adaptive.

2:30 PM - 2:45 PM (15 mins)
Evolution of the nuclear pore complex
Mark Field, Jennifer Holden, Ludek Koreny, Samson Obado, Brian Chait, Michael Rout
University of Dundee, The Rockefeller University

The nuclear envelope represents a major innovation and one that separates the eukaryotic from prokaryotic cell type. Further, the nuclear envelope participates in many functions, and which include control of gene expression via transcriptional state changes, mRNA processing and mRNA export. We have been investigating the biology of the trypanosome nuclear envelope in order to understand the mechanisms that trypanosomes use to control expression, and how these relate to those used by higher eukaryotes. Recent evidence supports a role for nuclear pore complex proteins in the control of spindle organisation, critical to mitosis and chromosomal segregation. However, our data also suggests complex evolutionary trajectories, and which likely have important impact on how the nuclear envelope arose and how it functions in different lineages.

2:45 PM - 3:00 PM (15 mins)
Architecture and evolution of the trypanosome nuclear pore complex
Samson Obado, Marc Brillantes, Wenzhu Zang, Mark Field, Brian Chait and Michael Rout
The Rockefeller University, University of Dundee

Much of the core architecture of the eukaryotic cell was established over one billion years ago. However, many cellular systems possess lineage-specific features, and architectural and compositional variation of complexes and pathways is likely keyed to specific functional differences. The nuclear pore complex (NPC) is responsible for many processes including nucleocytoplasmic transport, interactions with the nuclear lamina and mRNA processing. NPC structure and composition are best documented in *Saccharomyces cerevisiae* and mammals. We exploited trypanosomes to investigate NPC evolution at the level of conservation of protein-protein interactions and composition, allowing us to unambiguously assign NPC components to specific substructures. Thus the NPC structural scaffold is generally conserved, albeit with lineage-specific elements. However, there is significant variation in pore membrane proteins and an absence of critical components involved in mRNA export in fungi and animals. Identification of these lineage-specific features within the trypanosome NPC significantly advances our understanding of the mechanisms of nuclear transport, gene expression and of the evolution of the nucleus.

3:00 PM - 3:15 PM (15 mins)

Identification of a novel adaptin-related coat complex *

Alexander Schlacht[2], Jennifer Hirst[1], John P. Norcott[3], David Traynor[4], Gareth Bloomfield[4], Robin Antrobus[1], Robert R. Kay[4], Joel B. Dacks[2], Margaret S. Robinson[1]

[1.]University of Cambridge, Cambridge Institute for Medical Research, Cambridge, UK; [2.]Department of Cell Biology, University of Alberta, Edmonton, Canada; [3.]University of Cambridge, Department of Engineering, Cambridge, UK; [4.]MRC Laboratory of Molecular Biology, Cambridge, UK

We identified a novel complex related to the APs and COPI coats. Functional analysis in *Dictyostelium* indicates that this complex is a heterohexamer, with two components containing the typical protocoatomer membrane deformation architecture. We identified a complete complex in representatives from three eukaryotic supergroups, and partial complexes in each of the major lineages of eukaryotes, indicating its presence the Last Eukaryotic Common Ancestor, followed by multiple instances of secondary loss. Interestingly, a remnant of the medium subunit has been retained in animals and fungi, producing the origin of a mu homology domain important for endocytosis. The addition of this novel complex to the repertoire of ancient heterotetrameric complexes allows us to deduce an evolutionary path from the earliest origins of the heterotetramer-protocoatomer coat to its multiple manifestations in modern organisms.

3:15 PM - 3:30 PM (15 mins)

NUP-2, a second component of the trypanosome nucleoskeleton *

Luke Maishman[1], Obado, Samson[2]; Alsford, Sam[3]; Bart, Jean-Mathieu[4]; Navarro, Miguel[4]; Horn, David[3]; Chait, Brian[2]; Rout, Michael[2]; Field, Mark[5]

[1.]University of Cambridge; [2.]The Rockefeller University; [3.]London School of Hygiene and Tropical Medicine; [4.]Institute of Parasitology and Biomedicine "López-Neyra"); [5.]University of Dundee

Nuclear lamins are required for a diverse array of cellular functions, from chromatin organisation and transcriptional regulation to nuclear structure and physical support for the cytoskeleton. However, lamin homologs are restricted to animals and amoeba, and absent from all other taxa. NUP-1 is a 450 kDa coiled coiled protein at the nuclear periphery in African trypanosomes with functions highly similar to nuclear lamins, encompassing nuclear structure, chromatin organisation and transcriptional regulation, suggesting that it is a component of the trypanosome nuclear lamina. Recently, using cryomilling, immunoaffinity isolation and ESI MS/MS we identified NUP-2, which is also a coiled-coil protein. Both NUP-1 and NUP-2 interact with the nuclear pore complex. NUP-2 locates, in a punctuate distribution, to the nuclear periphery throughout the cell cycle, in close proximity to NUP-1, the NPC and the telomeric ends of the chromosomes. RNAi-mediated silencing of NUP-2 leads to defects in proliferation, gross nuclear structure, nuclear envelope ultrastructure, NPC arrangement and chromatin organisation. Further, transcription at telomeric-proximal sites, including VSG expression sites, is altered in NUP-2 depleted cells, suggesting a role in transcriptional regulation. However, unlike NUP-1, NUP-2 depletion did not lead to an increase in VSG switching. Finally, the location of NUP-1 is affected in NUP-2 depleted cells, and likewise NUP-2 becomes disorganised when NUP-1 is depleted, implying that these are interdependent protein networks. We suggest that NUP-2 is a second component of the highly unusual trypanosomatid nuclear lamina.

Session D5 (RCA) - Helminths Co-infections and transmission dynamics

Chair: Andrea Graham, Princeton University

2:00 PM - 2:30 PM (30 mins)

Dynamics of helminth-microparasite co-infections, from pairwise interactions in the lab to realistic complexity in the field

Andrea Graham
Department of Ecology & Evolutionary Biology, Princeton University, USA

Co-infection of a host by helminths and microparasites (including protozoa, fungi, bacteria and/or viruses) is widely reported in natural systems. Such co-infections may affect both the individual host (e.g., if helminths affect microparasite load and/or severity of microparasite-associated disease) and the host population (e.g., if helminth-induced changes to within-host dynamics also affect transmission potential). To what extent can we understand and even predict the outcomes of such interactions? Once we can predict outcomes of pairwise co-infections in the lab, how well will this extend to multi-species complexity in the field? Experimental evidence from laboratory systems reveals that consequences of pairwise co-infection may indeed successfully be predicted from first principles. I will discuss a detailed example of how helminth-induced changes in immune activity and in availability of target cells for microparasites together enable prediction of co-infection dynamics during pairwise interactions between parasite species (one nematode and one apicomplexan). I will then discuss the extension of this predictive framework to a more realistic level of system complexity, drawing upon experimental and observational data from several wild systems. I argue that these steps, though challenging, are necessary for effective management of host health and parasite transmission in co-infected natural populations.

2:30 PM - 2:45 PM (15 mins)

Effects of parasite interactions on the survival of a wild rodent

Godefroy Devevey, Andy Fenton, Sarah Knowles, Owen Petchey & Amy Pedersen
University of Edinburgh, University of Liverpool

Parasites are usually assumed to have a negative effect on the health and fitness of their host. However, quantifying the effects of chronic infections on host health can be challenging, especially in the wild. In addition, since hosts are frequently co-infected by several parasites, predicting the effect of the removal of a single parasite can be difficult. In a series of field experiments in which we used the anthelmintic Ivermectin to reduce nematode burdens, we show that the effects of nematodes on the wood mouse *Apodemus sylvaticus* are highly context dependent. We found evidence of a negative within-host interaction between gastrointestinal parasites, such that the presence of the nematode *Heligmosomoides polygyrus* reduced the proliferation of the coccidial parasite *Eimeria hungaryensis*. In a second experiment, we show that *H. polygyrus* infection reduces the survival of wild wood mice. However, the negative effects of *H. polygyrus* infection were mitigated in mice coinfected with *E. hungaryensis*. These co-infected individuals suffered from antihelminthic treatment with a reduction in lifespan. Overall we demonstrate that the impact of one parasite species is highly context dependent, varying from being detrimental to beneficial, depending on its interactions with other, co-infecting parasites. These experiments therefore show how drug treatment can have unintended consequences on the host, suggesting that treatments should be tailored to individual circumstances.

2:45 PM - 3:00 PM (15 mins)

Female host sex-biased parasitism with *Mastophorus muris* in wild bank voles (*Myodes glareolus*). *

Maciej Grzybek[1], Jerzy M. Behnke[3], Anna Bajer[2], Mohammed Alsarraf[2].
[1.] Department of Parasitology and Invasive Disease, Faculty of Veterinary Medicine, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland; [2.] Department of Parasitology, Institute of Zoology, Faculty of Biology, University of Warsaw,1 Miecznikowa Street, 02-096, Warsaw, Poland; [3.] School of Life Sciences, University of Nottingham, University Park Campus, NG7 2RD, Nottingham, UK.

In populations of vertebrate hosts, the prevalence and abundance of helminth parasites often differ between the sexes, and the bias is usually in favour of males showing higher prevalence and more intense infections. Sex-specific bias of helminth infections has been attributed to sex differences in host traits, including sex hormones and their influence on

the host immune response and hence capacity to resist infection, and also to differences in the behaviour and ecology of the two sexes leading to different levels of exposure to the infective stages of helminths. Bank voles are one of the most common rodent species in Polish woodlands. Their parasitic fauna, behaviour and genetics have been described comprehensively over the last decade. Therefore, bank voles represent a good model to study relationships between host traits and parasite infection strategies. Here, contrary to the more usually observed male sex-biased trends, we report female sex-biased parasitism with *Mastophorus muris* in Polish bank vole populations studied over an 11 year period. We took into consideration all the intrinsic and extrinsic factors that were quantified and focused particularly on pregnancy and lactation status.

3:00 PM - 3:15 PM (15 mins)

Costs of resistance and costs of infections on the fitness of the mosquito *Aedes aegypti* infected with the filarial nematode *Brugia malayi* *

Cristina Ariani, Katherine Short, Sophia Smith, Punita Juneja and Francis M. Jiggins
University of Cambridge, Department of Genetics, Downing Street, CB2 3EH, Cambridge, UK

The spread of genes that confer insect's resistance to parasites can be directly associated to the fitness costs it may impose. In the presence of the parasite, it is likely that resistant individuals will be favoured over susceptible, as the first's immune system is able to kill the parasite. But in the absence of the parasite a resistant genotype will only be favoured if having a mounted immune system is not very costly. We used the mosquito *Aedes aegypti* and the filarial nematode *Brugia malayi* as a model system to investigate costs of resistance and costs of infection on the fitness of the mosquito host. Because resistance is controlled by a single locus, we used a cross to generate resistant and susceptible mosquitoes that differ only in this region of the genome. We fed each cross with infected and uninfected blood. We analysed longevity, fecundity and the feeding behaviour (if the parasite was manipulating the mosquito's attractiveness to blood) of both groups. Our results showed that susceptible infected individuals died faster than other treatments, showing a cost of infection. We did not detect any differences in fecundity between treatments. The feeding behaviour of the resistant cross differed to the susceptible cross regardless of the infection status, suggesting that the parasite is not manipulating the host's feeding behaviour. We conclude that there is little, if any, cost to mosquitoes of carrying the resistant gene under laboratory conditions.

3:15 PM - 3:30 PM (15 mins)

Onchocerciasis transmission in Ghana: the effect of simuliid cytospecies and host blood-meal choice

Poppy Lamberton[1], Robert A Cheke[1,2], Rory J Post[3,4], Peter Winskill[1], J Lee Crainey[4], Daniel A Boakye[5], Mike Y Osei-Atweneboana[6], Iñaki Tirados[2], Michael D Wilson[5], Anthony Tetteh-Kumah[7], Sampson Oto'o[5], María-Gloria Basañez[1]
[1.]Department of Infectious Disease Epidemiology, School of Public Health, Imperial College London, St Mary's Campus, London, W2 1PG, UK; [2.]Natural Resources Institute, University of Greenwich at Medway, Central Avenue, Chatham Maritime, Kent, ME4 4TB, UK; [3.]School of Natural Sciences and Psychology, Liverpool John Moores University, Byrom Street, Liverpool L3 3AH, UK; [4.]Disease Control Department, London School of Hygiene and Tropical Medicine, Keppel Street, London, WC1E 7HT, UK; [5.]Noguchi Memorial Institute for Medical Research, University of Ghana, Legon, Accra, PO Box LG581, Ghana; [6.]Water Research Institute, Council for Scientific and Industrial Research, Accra, PO Box M32, Ghana; [7.]Ghana Health Service, Private Mail Bag, Ministries, Accra, Ghana

The proportion of blood-meals taken by vectors on humans is important for understanding the epidemiology of diseases such as onchocerciasis, whose only definitive hosts are humans, but whose vectors are known to feed on a range of hosts. Seven study sites in four regions of Ghana were visited from 2009 to 2011 in both rainy and dry seasons. A total of 17,300 blackflies were collected (98.8% *Simulium damnosum* s.l.), 6,142 by vector collectors; 2,207 in a man-baited gazebo, 1,567 in a cow-baited gazebo, 7,212 on Bellec traps and 172 in Monk's Wood light traps. Flies were stored for molecular and morphological identification of fly species and blood-meal origin. A total of 11,229 abdomens of parous females were tested, with blood-meals successfully amplified in 3,820. Host species included human (3,053), porcine (343), bovine (211), ovine (18), canine (14), taurine (9) and 1,253 unidentified. A single host species was identified in 76% of the flies (n = 2,902), of which 76% were human (n = 2,207), but up to 4 different hosts were recorded in a few flies. *S. soubrense* Beffa form (94% human) and S. squamosum (88%) showed greater anthropophagy than *S. damnosum* s.s./*sirbanum* (74%), *S. sanctipauli* (74%) and *S. yahense* (61%). This was supported in the savannah regions by higher proportions of *S. damnosum* s.s./*sirbanum* caught using cattle bait, in comparison to all other cytospecies. Our results show that host choice varies between cytospecies, and may be affected by vector and/or host density with epidemiological relevance for vector-borne disease models.

Session E5 (RCU) - Ecology - Aquatic Parasitology & Sustainable Fisheries, in memory of Professor Angela Davie

Chair: Rachel Norman, University of Stirling

This session is dedicated to the memory of Professor Angela Davies (Russell), one of the world's experts on the diseases of cold-blooded animals, whose research particularly focused on the blood parasites of fish and reptiles. She was an Emeritus Professor at Kingston University and worked closely with colleagues at the University of Johannesburg and North-West University, South Africa. She was awarded the prestigious Elsdon-Dew Medal from the Parasitological Society of Southern Africa for her research and was one of the very few UK parasitologists to receive this honour. She leaves a legacy of over a hundred publications, several new species descriptions and many qualified parasitologists in the UK and South Africa who benefited from her immense knowledge, patience and kindness.

2:00 PM - 2:30 PM (30 mins)
Pathogen control in aquatic systems: A national challenge
Nick Taylor
Centre for Environment, Fisheries and Aquaculture Science (Cefas), Barrack Road, Weymouth, DT5 2ES

Aquatic systems provide ecologically diverse and economically important resources. In addition to providing important commercial and recreational fisheries, these systems are also utilised to produce aquatic animals for the ornamental, food and recreational fisheries sectors. The diversity of industries dependent on aquatic systems leads to complex interactions between sectors that make controlling, reducing impact, and stopping the introduction and spread of pathogens at the national level challenging. Nationally, comprehensive records of aquaculture sites and recreational fisheries and moments of fish between them are held, however, in comparison little is known regarding the distribution of wild fish species or the holding and dissemination of ornamental fish intended as companion animals. Understanding the interactions between these companion animals, wild populations and aquaculture production businesses is of great importance. This presentation describes efforts to prevent the introduction of pathogens of national importance to the UK, improve the efficiency of our surveillance programmes, contain and control introduced pathogens, and reduce their impact to aquaculture systems, fisheries and wild fish stocks.

2:30 PM - 2:45 PM (15 mins)
Parasitic or not? Symbiotic branchiobdellids (Annelida: *Clitellata*) on invasive signal crayfish (*Pacifastacus leniusculus*)
Jo James[1], Jo Cable[1], Graham Richardson[1], Andy Mackie[2]
[1.] School of Biosciences, Cardiff University, Cardiff, CF10 3AX, UK; [2.] Department of Biodiversity & Systematic Biology, National Museum Wales, Cathays Park, Cardiff CF10 3NP, UK.

Biological invasions provide an important pathway for the spread of parasites, potentially facilitating parasite recruitment to novel hosts. Here, we document the first record of two non-native ecto-symbiotic branchiobdellid worms, *Xironogiton victoriensis* and *Cambarus* sp., from invasive signal crayfish (*Pacifastacus leniusculus*) in Wales. In the invaded river, the branchiobdellids were abundant (76% and 70% prevalence with 39.5 and 2.8 mean intensity for *X. victoriensis* and *Cambarus* sp. respectively), with a positive correlation for both species between crayfish size and infection intensity. Co-infections were common, however the branchiodellids exhibited microhabitat segregation on the host with *X. victoriensis* being found mainly on the chelae and the *Cambarus* sp. on the carapace. Previous data suggests that the relationship between crayfish and branchiobdellids is dependent upon worm species, and can vary from mutualism to parasitism. Our laboratory experiments revealed that although *X. victoriensis* infection did not significantly affect crayfish growth rate, infected animals were significantly less aggressive, and so competitively subordinate, to uninfected individuals. Through reducing their competitive ability, we conclude that *X. victoriensis* is parasitic on signal crayfish. Supressed aggressively of signal crayfish is predicted to decrease their impacts on other aquatic organisms, therefore, *X. victoriensis* has the potential to serve as a bio-control agent for these invaders.

2:45 PM - 3:00 PM (15 mins)

Why does maternal food matter? Maternal effects on body size modify disease resistance in *Daphnia magna* offspring.

Jennie Garbutt, Tom J. Little

Institute of Evolutionary Biology, The University of Edinburgh, Kings Buildings, Ashworth Laboratories, West Mains Road, Edinburgh, EH9 3JT

Maternal effects triggered by changes in the environment can influence the outcome of offspring-parasite interactions, with profound fitness consequences for the host and parasite. Outside of the classic example of antibody transfer in vertebrates, proximate mechanisms have been little studied, and thus the adaptive significance of maternal effects on infection is not well resolved. We sought to determine why food-stressed mothers give birth to offspring that show a low rate of infection when the crustacean *Daphnia magna* is exposed to a bacterial pathogen. In this system the more-resistant offspring of food-stressed mothers are larger at birth and feed at a lower rate: reduced disease resistance likely results because offspring ingest fewer bacterial spores, or because their larger size allows for greater immune investment. To distinguish between these theories we measured body size, feeding rate and susceptibility in the same individuals and were able to show that body size is the primary mechanism causing altered susceptibility: larger Daphnia were less likely to become infected. In addition to maternal food availability many ecological factors (e.g. size-selective predation) influence the size structure of host populations, and thus our results highlight a broad mechanism by which ecological context can affect disease epidemiology.

3:00 PM - 3:15 PM (15 mins)

Changes in parasite within-host dynamics and estimates of genetic variation over time *

Melanie Clerc[1, 2], Dieter Ebert[1], Matthew D. Hall[1, 3]

[1.]Zoological Institute, University of Basel, Vesalgasse 1, Basel, CH-4051, Switzerland: [2.]Institute of Evolutionary Biology, University of Edinburgh, Ashworth Labs, West Mains Road, Edinburgh EH9 3JT, UK; [3.]School of Biological Sciences,Clayton Campus, Monash University, Melbourne 3800, Australia

Disease is a continuous and dynamic process, including distinct host and parasite specific phases. Over the course of infection, interaction patterns between host and parasite can change, with potential consequences for the expression of disease related traits and virulence. However, the full magnitude of variation in disease dynamics has rarely been addressed so far, as disease life history traits have typically been measured at one or few moments during infection. Therefore, we characterised the within-host dynamics of the castrating bacterial parasite *Pasteuria ramose*, which infects the crustacean water flea *Daphnia magna*, over an entire infection period on a fine time scale. We found high levels of variation for host castration, parasite transmission stage production and parasite-induced gigantism. Differences in castration efficiency among parasite clones was structured over time and corresponded with parasite spore production, which accounted for an increase in genetic variation for virulence and parasite fitness with infection age. We discuss the evolutionary consequences of this finding with regard to natural selection acting on specific ages of infection and the mechanism underlying the maintenance of castration efficiency. Our results demonstrate that infection-age-specific estimates of host and parasite fitness components and hence elucidating within-host dynamics can shed light into the selective forces that shape infection strategies and the evolution of virulence.

3:15 PM - 3:30 PM (15 mins)

Parasites of Trinidadian guppies, *Poecilia reticulata*: evidence for sex- and age-specific trait-mediated indirect effects of predators *

Jessica Stephenson[1], Cock van Oosterhout[2], Ryan S. Mohammed[3], Joanne Cable[1]

[1.]School of Biosciences, Cardiff University, Cardiff, United Kingdom; [2.]School of Environmental Sciences, University of East Anglia, Norwich Research Park, Norwich, United Kingdom; [3.]Department of Life Sciences, The University of the West Indies, St. Augustine, Trinidad and Tobago

Predation pressure can alter the morphology, physiology, life history and behaviour of prey; this can change how surviving prey interact with parasites. These trait-mediated indirect effects may change in direction or intensity as animals grow or, in sexually dimorphic species, between the sexes. The Trinidadian guppy *Poecilia reticulata* presents a unique opportunity to examine these interactions; its behavioural ecology has been intensively studied in wild populations with well-characterised predator faunas. In these populations, sex and age-specific anti-predator responses have evolved that could affect the transmission rate of directly transmitted parasites such as *Gyrodactylus* spp. To test for evidence of predator-driven differences in infection, we collected 4715 guppies from 62 sites across Trinidad in 2003-

2009 and screened them for ectosymbionts including *Gyrodactylus*. A novel model-averaging analysis revealed that females were more likely to be infected with *Gyrodactylus* parasites than males, but only in populations with high predation pressure where, additionally, a greater proportion of guppies was infected. From small-scale studies we know that in such high-predation sites, females (but not males) tend to shoal, and this anti-predator behaviour facilitates parasite transmission. We propose that these recognised sex differences in shoaling could explain the observed sex difference in infection incidence. The infection rate of juveniles did not vary with predation regime, probably because juveniles face predation pressure from adults and therefore tend to shoal both in high- and low-predation sites. This represents the first evidence for age- and sex-specific trait-mediated indirect effects of predators on the probability of infection in prey.

Session F5 (RCJ) Careers - Careers Workshop

Chair: Emily Dawson & Luke Roberts

2:00 PM - 2:15 PM (15 mins)
Wellcome Trust fellowship schemes
Mike Turner
Wellcome Trust

Mike is the newly appointed Head of Infection and Immunobiology at the Wellcome Trust. His main interests are in parasite genetics and genomics, and he has worked on many aspects of parasite biology: transmission and population genetics, genomics, antigenic variation, developmental biology and immunology. Mike will talk through the core Wellcome Trust fellowship schemes available for newly-qualified and junior postdoctoral scientists, including the Sir Henry Wellcome and Dale Postdoctoral Fellowships. He will also talk about 'grantsmanship', particularly about the nature of hypothesis driven research and the difference between addressing a question and testing a hypothesis. More information about Wellcome Trust fellowship schemes is available at: http://www.wellcome.ac.uk/Funding/Biomedical-science/Funding-schemes/Fellowships/Basic-biomedical-fellowships/index.htm

2:15 PM - 2:30 PM (15 mins)
Life as a junior academic
Emily Adams
Liverpool School of Tropical Medicine

Emily finished her PhD from Bristol University in trypanosomiasis in 2008 and during her write-up had applied for a post-doc position at the Royal Tropical Institute in Amsterdam, Netherlands. Moving to the continent was a big step away from family and friends, but she benefitted a huge amount from working in a well-connected, industrious institute. In 2013 she started a joint-lectureship at the Liverpool School of Tropical Medicine and Warwick University. A year on and she is in the toils of junior academic life; grant applications, papers, reviews, supervision and teaching. It is a varied and enjoyable job with lots of travel, interesting colleagues and their inspiring work, but it is not without its hardships. Emily will speak further on the life as a junior academic - the highs and the lows!

2:30 PM - 2:45 PM (15 mins)
Career paths and the bizarre challenges along the way
Mike Leahy
Oxford University - TV presenter.

Mike has a BSc in Ecology and a DPhil from Oxford University in Virology and Molecular Biology. After completing his DPhil, he worked as a virologist and microbiologist, both of which involved an element of parasitology. Most of his research was conducted at Oxford University, although he also worked overseas for short periods. Mike gave up his career as a researcher to become a full-time TV presenter. His shows include BBC's Rough Science, Sky1's Invasion of the Bodyscratchers and National Geographic Channel's Bite Me. He currently has a new venture "The Zoo-Bus", works as public speaker with education and business, and is a travel health and adventure writer. As well as covering his career path, Mike will talk about the various bizarre challenges he has undertaken during his career, including using his body as bait to catch the notorious penis-invading candiru fish, infecting himself with a tapeworm and risking death by permanent erection at the hands (or rather fangs) of the Brazilian Wandering Spider.

2:45 PM - 3:00 PM (15 mins)
How and why you should pursue various career options
Leslie Drake
The Partnership for Child Development, Department of Infectious Disease Epidemiology, Imperial College London

Lesley gained her PhD in epidemiology/parasitology from Imperial College London (ICL) where she investigated the inter-relationship between parasites and their human hosts and how this might be exploited in the quest for more effective drug and vaccine design. However, since then she has pursued a highly successful career outside of the more conventional remit of basic research. Strongly committed to child development issues, Lesley has gone on to become the executive Director of the Partnership for Child Development (PCD) at ICL, which focuses on strengthening the evidence base on effective implementation of school health and nutrition programmes in support of governments in low and middle income countries. Lesley also maintains the role of deputy director for the London Centre for Neglected Tropical Disease Research. How and why she pursued these career options, and how her PhD training made this possible will be discussed during her presentation.

Session A6 (WRA) - Kinetoplastida - Interactions with vertebrate and arthropod hosts

Chair: Mark Carrington, University of Cambridge

4:00 PM - 4:30 PM (30 mins)

***Leishmania* proteophosphoglycans (PPGs): genetic analysis of a parasite mucin-like glycoprotein implicated in several key steps in the infectious cycle**

Stephen Beverley[1], Marcos dos Santos[1], David Sacks[2]

[1.]Dept. of Molecular Microbiology Washington University School of Medicine, St. Louis, USA; [2.]Laboratory of Parasitic Diseases, NIH, Bethesda, USA.

Phosphoglycans (PPG) are abundant mucin-like glycoproteins found on the surface and secreted by the trypanosomatid protozoan parasite *Leishmania*. PPG proteins range up to 2 MDa, mostly comprised of tandemly repeated 15-17 amino acid serine-rich motifs. The peptide repeats in turn are heavily modified by substituted phosphoglycan (PG) repeating units [Gal(β1, 4)-Man(α1)-PO4], first identified in the more abundant *Leishmania* lipophosphoglycan (LPG). Unlike LPG where the PG repeating units are attached to a GPI-core acceptor, PG repeats are attached to the PPG protein backbone through Man-PO4-Ser linkages. PPGs are secreted and thus less abundant than LPG on the cell surface. Studies from several other groups have shown that PPGs are expressed throughout the infectious cycle, and have been proposed to carry out several unique roles independent of LPG. In the sand fly vector, PPGs form a major part of the gel-like 'plug' that alters feeding behavior and parasite transmission. In the mammalian host, inoculation of the 'plug' and/or PPG expression in the phagolysosome may exacerbate parasite infectivity and pathology. In previous studies we used specifically LPG-deficient/PPG-replete mutants (such as lpg1-) to unambiguously assess the unique roles of LPG across the parasite infectious cycle. Here we tackled the challenge (2 loci, very large genes, cluster >100 kb) of getting a specifically ppg-null mutant through deletion of the PPG protein backbone. PPGs are encoded by 5 genes at two loci; PPG2 on chromosome 33 and a cluster of >100 kb encoding PPG[3-4-5-1] on chromosome 35. We were able to generate homozygous replacements of both loci independently (ppg-cluster -or ppg2-), and a double mutant lacking both the PPG cluster and PPG2 (ppg-null). Southern blot analysis suggests the true size of the PPG cluster to be 180 kb. Western blot analysis with anti-PG antisera was used to confirm the loss of PPG expression. The results of studies exploring the consequences of PPG ablation across the infectious cycle will be described

4:30 PM - 4:45 PM (15 mins)

Unzipping the barriers: how trypanosomes breach the tsetse peritrophic matrix *

Clair Rose[1], Rodrigo Belmonte[2,3], Stuart D Armstrong[4], Gemma Molyneux[1] Lee Haines[2], Michael Lehane[2], Jonathan Wastling[4] and Alvaro Acosta-Serrano[1,2]

Department of [1.]Parasitology and [2.]Vector Biology, Liverpool School of Tropical Medicine, Liverpool, UK, [4.]Institute for Infection Biology, University of Liverpool, Liverpool, UK; [3.]Current address: School of Biological Sciences, University of Aberdeen, Aberdeen, UK

To migrate and develop within their tsetse host, African trypanosomes must overcome the flies' peritrophic matrix (PM), an acellular gut-lining surrounding the blood-meal. PM crossing occurs at least once during parasite migration, the mechanism of which is not understood. We have used a combination of transmission electron microscopy (TEM), mass spectrometry (MS) and RNAi knockdown to better comprehend the molecular events surrounding PM penetration. TEM analysis shows the tsetse PM is composed of a three-layer structure, including an electron-dense zone in direct contact with the midgut lumen. Trypanosome crossing appears to occur in at least three steps: 1) Breaking and "unzipping" the electron-dense layer 2) tunnelling throughout the middle layer 3) access to the ectoperitrophic space from a point distal to that of entry. An MS-based proteomic analysis of isolated PM proteins submitted to either in-solution digestion or fractionated on 1D SDS-PAGE, followed by in-gel trypsinization, showed the Glossina morsitans PM is composed of nearly 300 proteins, the most abundant being the immunomodulator TsetseEP, and specific PM proteins, including novel peritrophins and peritrophin-like glycoproteins that are essential in maintaining PM architecture. Furthermore, a minimum of 27 proteins from the tsetse secondary endosymbiont, *Sodalis glossinidius*, were also identified, suggesting a close association with the PM. Preliminary RNAi silencing of a peritrophin, GmmPro2, leads to a significant increase in trypanosome infections, suggesting that perturbations in the PM architecture may facilitate parasite crossing. We are currently analysing by EM and fluorescence microscopy the possible structural changes of the PMs of knockdown flies.

4:45 PM - 5:00 PM (15 mins)

Both host and parasite genetic factors determine long-term tissue-specific infection dynamics in experimental chronic Chagas disease

Michael Lewis, Amanda Fortes Francisco, Martin C. Taylor, Michael A. Miles and John M. Kelly

Department of Pathogen Molecular Biology, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, Keppel Street, London, WC1E 7HT

The ability to establish persistent infection in a restricted set of 'permissive' tissues is a key immune evasion strategy used by the protozoan parasite *Trypanosoma cruzi*. However, the mechanism and pathological relevance of this tissue tropism in Chagas disease are poorly understood. Progress in this area has been hindered by a lack of sensitive methods to detect rare, focally-distributed parasites within tissues. Using a novel, highly sensitive bioluminescence imaging approach we studied the course of infection and chronic, tissue-specific distributions of two divergent *T. cruzi* strains, CLBR (TcVI) and JRcl4 (TcI), in three inbred mouse models: BALB/c, C57BL/6 and C3H/HeN. After an acute phase peak, parasite burdens in all six host-parasite combinations were efficiently controlled resulting in the establishment of a dynamic but stable chronic phase. Five months post-infection, tissue-specific parasite burdens were quantified by ex vivo bioluminescence imaging conducted immediately post-mortem. The gut was consistently found to be the major site of bioluminescent foci in all host-parasite combinations identifying this organ as the primary site of *T. cruzi* persistence. The heart, which is the most important site of Chagas disease pathology, and other organs were differentially associated with bioluminescence, dependent on both parasite strain and host genetic background. Histological analysis of cardiac inflammation and fibrosis allowed the association of heart-specific infection with markers of pathogenesis to be investigated. Taken together these data indicate that tissue tropism in long-term *T. cruzi* infections is determined by interaction of both host and parasite genetic factors.

5:00 PM - 5:15 PM (15 mins)

In search of anti-disease vaccine candidates for African trypanosomiasis: new insights into the protein composition of trypanosome-infected tsetse saliva

Alvaro Acosta-Serrano, Samïrah Perally, Christopher Williams, Ambalika Batra, Lourdes Duque, Lee Haines, Michael Lehane

Department of Parasitology and Department of Vector Biology, Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA

During the tsetse life cycle of all *Trypanosoma bruce*i sub-species, the parasite colonizes the fly's salivary glands (SG) to develop into the saliva-borne metacyclic infectious form. Interestingly, establishment of a SG trypanosome infection is accompanied by a down-regulation (~75%) in the overall expression of salivary proteins, which causes a feeding phenotype and helps disease spreading (Van Den Abbeele et al., 2010). Despite the importance of tsetse saliva in disease transmission, nothing is known about its composition (sialome) during a trypanosome infection. We reasoned that saliva from trypanosome-infected flies contain soluble factors that may be important for parasite transmission into the mammalian host. Using highly sensitive proteomic methodologies, we identify proteins found in the saliva of trypanosome-infected flies. Tsetse saliva was collected from *Trypanosoma brucei*-infected and parasite-naïve *Glossina morsitans* flies. Western blotting analysis using an antiserum against tsetse saliva showed that salivary proteins from infected flies are partially degraded. Following tryptic digest and LC-MS/MS analysis, salivary proteins were identified in naïve (128) and trypanosome-infected (267) sialomes. Furthermore, LC-MS/MS analysis also showed abundant trypanosome GPI-anchored glycoproteins in infected tsetse saliva, namely Brucei Alanine Rich Protein (BARP), Fam50 proteins (which are phylogenetically related to the BARP family (Jackson *et al.,* 2013)) and variant surface glycoproteins. Interestingly, tsetse saliva from either naïve or trypanosome-infected flies also contains factors from bacterial symbionts. Thus, trypanosome-infected tsetse saliva is a cocktail comprised of soluble tsetse, parasite and bacterial molecules, which may play pivotal roles during parasite transmission. These could potentially lead to novel anti-disease.

5:15 PM - 5:30 PM (15 mins)

Novel method for quantifying *Leishmania* metacyclics promastigotes delivered by sand fly bite.

Matthew Rogers, Emilie Giraud, Oihane Martin
London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT

Transmission of leishmaniasis results from regurgitation of parasites from the anterior midgut of the sand fly vector. Regurgitation is facilitated by the accumulation of promastigote secretory gel (PSG) that blocks the sand fly gut and manipulates bloodfeeding. Recently, real-time quantitative PCR (RTqPCR) has given us greater insight into the transmission process from individual sand flies, highlighting its variability and association with the intensity of the midgut infection. Here we describe the use of RTqPCR to determine the composition of the infectious dose by quantifying the proportion of metacyclics present through detection of SHERP (small hydrophilic endoplasmic reticulum-associated protein) transcripts. We show that expression of *Leishmania mexicana* and *Leishmania infantum* SHERP correlates with the proportion of metacyclics in both mouse skin and sand fly midguts. On the rare occasion that the composition of the infectious dose has been investigated by microscopy, a highly enriched population of metacyclics has been recorded. Using the metacyclic RTqPCR we: (i) corroborate this finding from infected sand fly bites, (ii) uncover unique aspects of the host's immune response to transmission and (iii) highlight novel interactions between *Leishmania* and the PSG prior to transmission. Finally, we discuss the potential role of this method to improve the screening of anti-leishmanial vaccines.

Session B6 (WRR) - Apicomplexa - Other Apicomplexa

Chair: Janet Cox-Singh, University of St Andrews

4:00 PM - 4:30 PM (30 mins)

Adaptive biology of the persistent human parasites *Plasmodium malariae, P. ovale curtisi* and *P. ovale wallikeri*

Colin Sutherland, Mary Oguike, Debbie Nolder, Peter L. Chiodini
London School of Hygiene and Tropical Medicine

Recent studies of *P. malariae* and *P. ovale* spp. infections in travellers returning to non-endemic countries from Africa have identified novel aspects of the biology of each of these parasites which enhance their persistence in the human host. At the same time, application of sensitive molecular detection methods in field studies have shown these three species to be far more common in Africa, Asia and the SW Pacific than previously acknowledged. Worryingly, there is some evidence that artemisinin combination therapy is not completely effective at eradicating the low density, sub-microscopic infections that are characteristic of all three species. Recent data on parasite biology and drug response patterns in vivo will be presented for each of these parasites.

4:30 PM - 4:45 PM (15 mins)

Structure, Evolution and Function of the Genome of the Emerging Human Pathogen *Babesia microti*.

Emmanuel Cornillot[1], Aprajita Garg[2], Amina Dassouli[2,3], Niseema Pachikara[2], Sahar Usmani-Brown[2], Anna Stein[2], William Zhao[2], Qi Su[4], Hanzel T. Gotia[4], Priti Kumari[4], Joshua Orvis[4], Ankit Dwivedi[1], Jonathan Crabtree[4], Kyle Tretina[4], Roger Frutos[1], Peter J. Krause[5], Claire Fraser[4], Stephane Delbecq[3], Joana Silva[4] and Choukri Ben Mamoun[2].
[1.] CPBS – UMR 5236, Montpellier, France & Institut de Biologie Computationnelle, Montpellier, France; [2.] Department of Internal Medicine, Section of Infectious Diseases, Yale School of Medicine, New Haven, Connecticut, USA; [3.] LBCM-EA4558, UFR Pharmacie, Université Montpellier 1, Montpellier, France; [4.] Institute for Genome Sciences, University of Maryland School of Medicine, Baltimore, Maryland, USA; [5.] Yale School of Public Health and Yale School of Medicine, New Haven, Connecticut, USA

Babesia microti is an apicomplexan intraerythrocytic parasite that causes human babesiosis, a malaria-like illness found worldwide and endemic in the United States. The parasite is transmitted to humans by the tick vector, *Ixodes scapularis*. It is also the most common transfusion-transmitted pathogen in the United States. In order to gain better understanding of the biology, pathogenesis and evolution of *B. microti* and to develop new diagnostic assays and therapies, we formed a consortium to establish comprehensive genomic resources to be available to the research community. Our goals include the generation of genome and transcriptome assemblies for multiple *B. microti* isolates, perform metabolic reconstruction and comparative functional analyses. The nuclear genome of the reference R1 strain was published in 2012; at 6.5Mb, it is the smallest known apicomplexan genome. It encodes approx. 3500 proteins, ~12% of which are species-specific. *B. microti* has a linear mitochondrial genome present in four different structural types and a 28 kb circular apicoplast genome with a gene organization reminiscent of its chloroplast ancestor. Phylogenetic analyses revealed that *B. microti* defines a new lineage in the apicomplexan phylum distinct from lineages encompassing *Plasmodium* sp, as well as other piroplasmida taxa such as *Theileria* and *Babesia bovis*. Metabolic reconstruction studies identified several pathways that could be targeted for development of target-specific therapies against *Babesia*. Current progress of the consortium includes the newly characterized apicoplast genome, the updated annotation of the nuclear genome, and estimates of genomic diversity in the species based on the sequencing of new clinical isolates.

4:45 PM - 5:00 PM (15 mins)

Whole Genome Amplification from clinical samples: Advances in *Cryptosporidium* genome sequencing from limited numbers of oocysts *

Jenna Alexander[1], Stephen J Hadfield[2], Guy Robinson[2], Kristin Elwin[2], Rachel M Chalmers[2], Justin A Pachebat[1]
[1.] Institute of Biological, Environmental and Rural Sciences, Aberystwyth University, UK; [2.] Cryptosporidium Reference Unit, Public Health Wales Microbiology, Singleton Hospital, Swansea, UK

The study of *Cryptosporidium* is hampered by an insufficient understanding of species sub-types and diversity. Whole

genome reads can give a wealth of information about infectivity, phylogeny, and host specificity, but to date only three species have been genome sequenced and published. Recent advances in next generation sequencing (NGS), and the use of the transposon based Illumina Nextera XT sequencing library preparation kit, has allowed us to characterise *Cryptospordium* genomic sequences directly from clinical samples from which over a nanogram of DNA can be extracted (See Hadfield et al., Pachebat et al., posters at this meeting). One limiting factor in the generation of sequence data directly from isolates is the small number of oocysts that can be recovered from clinical samples, often resulting in the extraction of sub-nanogram quantities of genomic DNA. Here we discuss the application of single molecule genomic DNA extraction methods to improve the recovery of genomic DNA from oocysts, and the efficiency of commercial available single cell whole genome amplification (WGA) kits to amplify a range of genomic DNA concentrations isolated from *Cryptosporidium parvum* oocysts. We also discuss NGS sequence characterisation of the WGA amplified DNA to determine the efficacy of the various WGA methods, and the application of this method to clinical isolates stored at the Cryptosporidium Reference Unit.

5:00 PM - 5:15 PM (15 mins)

***Plasmodium knowlesi* - another malaria parasite.**

Janet Cox-Singh
School of Medicine, University of St Andrews, Fife, Scotland.

Plasmodium knowlesi, a parasite adapted to long and pigtailed macaques, has entered the human population in Southeast Asia. Human cases of *P. knowlesi* malaria are predominantly zoonotic and occur across the region suggesting on-going multiple entry events. Patients with confirmed single-species *P. knowlesi* infections present with a spectrum of disease ranging from low parasitaemia and uncomplicated malaria to high parasitaemia with severe and fatal outcomes. As a zoonosis, the spectrum of disease and parasitaemia observed in human hosts may be predominantly mediated by parasite diversity and less likely to be influenced by pre-existing host immunity. Erythrocyte invasion by *Plasmodium* species is a multi-step process involving several gene families leading to recognition, reorientation and merozoite entry into permissive erythrocytes. The *P. knowlesi* genome has two members of the reticulocyte binding-like gene family, *P. knowlesi* normocyte binding protein (Pknbp)xa and Pknbpxb, that encode merozoite proteins involved in erythrocyte selection and invasion. Therefore, since parasitaemia is associated with disease severity, we tested the hypothesis that particular alleles of Pknbpxa and Pknbpxb have an invasion advantage. For this study polymorphic fragments of each locus were sequenced in 147 P. knowlesi patient isolates. Seventy-five (75) Pknbpxa and 51 Pknbpxb haplotypes were resolved with haplotype diversity 0.9729 (SD ± 0.007) and 0.922 (SD ±0.014) respectively. The isolates formed twelve Pknbpxa and two Pknbpxb haplotypes groups with two or three alleles per group. Analysis for allelic clustering with parasitaemia and other markers of disease progression identified putatively virulent Pknbpxa and Pknbpxb alleles. The results will be presented within the context of parasite diversity and virulence.

5:15 PM - 5:30 PM (15 mins)

The European Malaria Reagent Repository - the Rodent Malaria Collection.

Joanne Thompson, Sarah Reece, David R. Cavanagh, Alexandra Rowe, David Arnot, Nio McGill and Kim Wilson.
Institute of Immunology and Infection Research, Centre for Immunity, Infection and Evolution, University of Edinburgh, King's Buildings, University of Edinburgh, Edinburgh, EH9 3JT, UK

Over the last six decades, the rodent malaria parasites, *Plasmodium chabaudi, P. vinckei, P. yoelii* and *P. berghei*, have become key model systems in malaria research. All rodent malaria parasites used today derive from isolates of blood collected from wild-caught thicket rats in Cameroon, Central African Republic, Congo, Democratic Republic of the Congo and Nigeria between 1948 and 1974. Edinburgh houses more than seventy of these isolates as well as numerous original strains from other research groups. From these, more than a hundred genetically-distinct subspecies and strains have been cloned and characterized. With the addition of distinct strains arising from genetic crosses and selection for drug-resistance, the Edinburgh collection has grown to become the largest collection of rodent malaria parasite strains available. We have created the "European Malaria Reagent Repository" which is dedicated to the development and curation of these reagents, and are now ready to launch this resource to the malaria community. The repository will replenish and renew rodent malaria parasite isolates and strains, comprehensively annotate them in an online database, and increase their accessibility to academic and commercial researchers.

Session C6 (WRL) - Vectors and Parasites in the Middle East - Vectors and Parasites in the Middle East I

Chair: Ashraf Mohamed Ahmed Ali, King Saud University

4:00 PM - 4:30 PM (30 mins)

Malaria in Saudi Arabia, the current situation.

Saeed Al-Harthi

Department of Medical Parasitology, Faculty of Medicine, Umm Al-Qura University, P.O. Box 13955, Makkah, Saudi Arabia. email: sasharthi@uqu.edu.sa

About 8% of Saudi population lives in areas of malaria transmission where *Plasmodium falciparum* accounts for almost all indigenous cases. Several Anopheles species have been identified as potential malaria vectors. Currently, over 98% of reported cases are imported from neighbouring Yemen and Asian/African endemic countries by emigrants and pilgrims. In recent history, the worst malaria outbreak happened in 1998 with more than 35000 locally transmitted cases. In year 2010, 1941 malaria cases were registered, among which only 29 were local infections. Malaria control programme started in 1940s. Eastern and central areas were declared malaria free by 1970s, but south-western provinces are still endemic. KSA is on track of meeting the Roll Back Malaria and World Health Assembly target of 75% reduction in malaria cases by 2015. Vector control relied on DDT followed by Dieldrin insecticides mass spraying. Pyrethroid Reslin and Fenitrothion were employed in persistent foci by mid 1980s. Larval control measures were introduced in 1960s as mosquito DDT-resistance increased. Although, demands for more environmentally friendly control measures are growing, little is known on current vector insecticide-resistance situation to permit targeted strategies. Free diagnosis and treatment are provided to patients. Malaria treatment follows worldwide tendencies to overcome parasites drug-resistance spread. Reports of Chloroquine resistance started appearing in 1990s. More recently, parasites resistance to pyrimethamine /sulfadoxine was reported. Since 2007, combinations of pyrimethamine/sulfadoxine/ artesunate were adopted as first line and lumefantrine/artemether as second line treatment for uncomplicated falciparum-malaria. For severe falciparum-malaria quinine and artesunate are used. Chloroquine and primaquine combination is used for vivax-malaria treatment.

4:30 PM - 4:45 PM (15 mins)

Hydatidosis and Echinococcosis in Gaza strip

Adnan Al-Hindi[1], Philip Craig[2], Boufana Belgees[2], Judith Smith[2], Freya van Kesteren[2] Mwangi Judy[2]

[1.] Islamic University of Gaza, Faculty of Health Sciences, Medical Laboratory Sciences Department P.O.Box 108, Gaza strip, Palestine; [2.] Cestode Zoonoses Research group, School of Environment and Life Sciences, University of Salford, Greater Manchester M5 4WT, UK

Echinococcosis is a disease caused a causative organism, the dog tapeworm *Echinococcus granulosus*, is transmitted cyclically between cannines and numerous herbivorous livestock animals, which can serve as intermediate hosts (including sheep's and rodents). To study the situation of Hydatidosis and Echinococcosis in Gaza Strip, about 37 DNA samples were brought from Gaza, which were extracted from faecal samples of necropsed dogs, and samples examined for *Ecchinococcus granulosus* (Cestode group). DNA Stool Mini Kit was used according to the instructions of manufacturers. A total of 19 sera samples were collected from farmers in Gaza strip and brought Salford University (Cestode group) to be examined for two antigens: Ag B and Hydatid cyst Fluid (HCF). It was found that 2/37 (5.4%) of dogs were infected with *Echinococcus granulosus* using copro-DNA in Gaza Strip. It was found that one positive case with Hydatid cyst disease from Gaza. We conclude that there is existence of *Ecchinococcus granulosus* among dogs in Gaza. The studied dogs were roaming and considered as stray dogs which defecate in streets, farms and constitute a high risk for the local community. It is recommended that documentation of human cases with hydatidosis is made in the Ministry of Health and that improved availability of Hydatid cyst disease test diagnosis in Gaza Strip is achieved.

4:45 PM - 5:00 PM (15 mins)

Morphological and Phylogenetic analysis of *Serrasentis sagittifer* (Acanthocephala: *Rhadinorhynchidae*) isolated from the Gilthead Sea bream *Sparus aurata* (*Sparidae*) , Red Sea, Egypt

Rewaida Abdel-Gaber[1], Abdel Rahman Bashtar[1], Saleh Al Quraishy[2]

[1.] Zoology Department, Faculty of Science, Cairo University, Cairo, Egypt; [2.] Zoology Department, College of Science, King Saud University, Riyadh, Saudi Arabia

Seventy specimens of the gilthead sea bream *Sparus aurata* of the Red Sea were collected during the period from March to November 2013; they were dissected and examined for parasitic acanthocephalans. Only Forty (57.14%) specimens were found to be naturally infected with *Serrasentis sagittifer* belonging to family Rhadinorhynchidae. The infection was recorded in the intestine, pyloric ceca, and the external surfaces of some internal organs of the infected fish. Light and scanning electron microscopy revealed that the adult worm was elongated (with broad anterior and narrow posterior ends) measured 6.9-8.6 (7.6±0.2) x 0.57-0.73 (0.63±0.02) mm for male and 10.2-12.1 (11.5±0.2) x 0.71-0.82 (0.76±0.02) mm for female. Proboscis was long and cylindrical with a length of 0.97-1.6 (1.2±0.2) mm for male and 1.12-1.17 (1.14±0.02) mm for female. It was covered with numerous uniform spines arranged longitudinally as 9-11 rows each equipped by 15-18 spines. Proboscis is followed by a short spineless neck region followed by the body proper which is supported by multiple combs of spines (16-20) on its ventral surface. Molecular analysis of the parasite 18s rDNA demonstrated a close identity (>83%) between the present acanthocephalan and other previously described species within class Palaeacanthocephala with a close identity 98% with the previously recorded *Serrasentis sagittifier* (Acc. No. JX014227). So according to the records of morphological and molecular analyses, the present parasite is classified as *S. sagittifier* belonging to class Palaeacanthocephala and family Rhadinorhynchidae with a new host record from the gilthead sea bream *Sparus aurata* of the Red Sea.

5:00 PM - 5:15 PM (15 mins)

Kudoa parasites infecting oocytes : characterization of new species arguing in favor of the induction of a xenoma-like structure

Lamjed Mansour
Department of Zoology, College of Science, King Saud University PO. Box: 2455, Riyadh, 11451, Saudi Arabia

Species belonging to the genus Kudoa (Meglitsch, 1947), are typically histozoic myxosporea infecting mainly marine fish. They count around 85 species, mainly infecting somatic muscle tissues. Few species were reported in internal organs as heart, gills, fins, intestines, bladders... In gonad , until recently, only one species, *Kudoa ovivora* (Swearer and Robertson, 1999) infecting oocytes of 7 labroid species was reported in in the Caribbean coast of Panama. A second species, *K. azevedoi* (Mansour et al; 2013) has just been reported within oocytes of the Atlantic horse mackerel *Trachurus trachurus* off Tunisian coasts. We report a third species infecting oocytes of the Indian Mackerel, *Rastrelliger kanagurta* in Saudi Arabia Coasts. Characterization based on structural, ultrastructural, and molecular data state for the creation of a new species in the genus *Kudoa*. We notice that based on the phylogentic analysis a strong cluster is formed by the three *Kudoa* infecting oocytes. The main important information is that these three kudoa parasites induce hypertrophy of the infected oocytes. Morphological and histological analysis show that hypertrophied oocytes are filled with mature spores while the yolk globules, the nucleus and the membrane of the oocyte are almost normal and not destroyed. This structure where the host cell is hypertrophied with parasites proliferating inside resembled xenomas interface observed in microsporidia infecting fish and in few other intracellular microscopic parasites

5:15 PM - 5:30 PM (15 mins) (Delegate unable to attend at last Miniute)

Molecular characterization of *Stictodora tridactyla* (Digenea: Heterophyidae) using ITS1 and mtCO1 sequences in Kuwait

Wafa Al-Kandari, Suzanne A. Al-Bustan1, Majed Alnaqeeb and Asha Isaac.
Kuwait University, Kuwait

Abstract withdrawn

Session D6 (RCA) - Helminths - Experimental Immunology - Mini Symposium

Chair: Richard Grencis/Andrew MacDonald, Manchester University

4:00 PM - 4:20 PM (20 mins)
Mark Wilson
NIMR, London

4:20 PM - 4:40 PM (20 mins)
Rick Maizels
Edinburgh

4:40 PM - 5:00 PM (20 mins)
Richard Grencis,
Manchester University

5:00 PM - 5:20 PM (20 mins)
Andrew MacDonald
Manchester University

5:20 PM - 5:30 PM (10 mins)
Discussion

Session E6 (RCU) - Ecology - Global Weirding, invasive species and parasitism

Chair: Jo Cable, Cardiff University

4:00 PM - 4:30 PM (30 mins)
Predicting the global emergence and spread of zoonotic infectious diseases
Kate Jones
Centre for Biodiversity and Environment Research University College London

There is growing interest in the role of ecosystems and the goods and services they provide in governing human health and well-being. In particular, intact habitats may act to regulate diseases emerging into human populations. If true, this powerful idea would provide a critical argument for the protection of biodiversity because global emerging infectious diseases are a huge burden to human health and economies. However supporting evidence for this link is contradictory and controversial. Empirical statistical analyses of human infectious diseases has been successful in understanding the spatial environmental correlates of initial outbreaks but a mechanistic understanding of these patterns is lacking at a global scale. Here I present some latest research attempting to merge both empirical and mechanistic approaches at a global scale using data from a poorly understood disease, Lassa fever. I show that macro-scale environmental conditions play an important role in driving the spatial extent of Lassa virus and that any future habitat change to agricultural systems may facilitate a range expansion. I also expand this approach to model zoonotic diseases more generally and examine the impact of future environmental and habitat change on the emergence and spread of zoonotic diseases.

4:30 PM - 4:45 PM (15 mins)
Predicting the effects of climate change on *Schistosoma mansoni* transmission in East Africa: A mathematical modelling study *
Nicky McCreesh, Nicky McCreesh and Mark Booth
School of Medicine, Pharmacy and Health, Durham University Wolfson Research Institute, Durham University Queen's Campus, University Boulevard, Thornaby, Stockton on Tees TS17 6BH

More than 200 million people are estimated to be infected with schistosomiasis. Both the intermediate host aquatic snail species and the Schistosoma parasite are sensitive to a wide range of different environmental factors, including temperature, and will therefore be affected by climate change. I have developed an agent-based model of schistosomiasis and water temperature. Snails, schistosome worms, cercariae and miracidia are represented as agents. Birth, development, mortality and transmission rates for the snails and parasites are temperature dependent. The model has been parameterised using experimental and field data from *S. mansoni* and *Biomphalaria pfeifferi* snails, and run using daily minimum and maximum temperatures from climate projections for East Africa between 2006 and 2076. Eight scenarios, representing different types of snail habitat, have been run for each of three different climate projections. Risk maps for mean schistosome transmission in 2006-2016 generated from model output fit well to current data on where schistosomiasis transmission is known to occur. In some areas, there is agreement between model scenarios that temperatures will become suitable for increased or decreased schistosomiasis transmission over the next 20 or 50 years, or that there will be little or no change in risk as a result of increasing temperatures. In other areas, the type of habitat may play an important role in determining the effects of increasing temperatures on schistosomiasis. The model output risk maps highlight areas at risk of epidemics, which should be monitored over coming years and decades.

4:45 PM - 5:00 PM (15 mins)
Distribution and abundance of Ixodid ticks as vectors of disease in Northern Ireland. *
Jonathan Lappin, N Marks, A Maule
Institute for Global food security MBC 97 Lisburn Road Belfast Co.Antrim N.Ireland BT9 7BL

This investigation examines the abundance and distribution of ixodid ticks throughout Northern Ireland (N.I.) and monitors the status of tick-borne diseases (TBDs). Our data indicate that there has been an increase in tick distribution and abundance in recent years. Increased tick density was confirmed through pitfall trapping and flagging at 15 forests in N.I.. Areas with high precipitation and humidity readings had higher tick numbers. Tick populations exhibited a bimodal

distribution peaking in June and again in September. A clear correlation between tick numbers and deer presence in forested areas was also noted as well as a correlation between tick density and TBDs in livestock, e.g. Babesiosis. It was also shown that forest type had a significant influence on tick abundance. Numerous tick species have been identified in N.I. the most common of which was *Ixodes ricinus* being recorded on deer, agricultural animals and domestic pets. Other species found include, *Ixodes trianguliceps* on rodents and foxes, *Ixodes hexagonous* on hedgehogs and *Ixodes canisuga* on badgers, foxes and dogs. This shows a diversity of species and hosts utilised by ticks to complete their life cycle and a greater range of reservoir hosts for the numerous pathogens ticks can carry.

5:00 PM - 5:15 PM (15 mins)

Seasonal dynamics and long term trends in a host-parasite community *

Caroline Millins[1], James Grecian[1], Bob Furness[1], Lucy Gilbert[2], Matt Denwood[1], Roman Biek[1]

[1.] Institute of Biodiversity, Animal Health & Comparative Medicine & Boyd Orr Centre, University of Glasgow; [2.] James Hutton Institute, Aberdeen

Environmental changes including climate warming and land management change are likely to affect host-parasite communities, with implications for disease spread and persistence in the host species. Here, we use a 17-year data set of passerine birds to test for long-term trends in the prevalence of the tick *Ixodes ricinus*, an important vector of several zoonotic and livestock pathogens in Europe. We investigated whether chaffinch and blackbird species exhibit the same temporal and seasonal trends in parasite burden with immature stages of *I. ricinus*, and whether sources of heterogeneity in tick burden due to sex or age differences were consistent among these species. Generalised linear mixed models were used to make statistically valid inference on all parameters. Seasonal and long term trends in tick burden were very similar in both chaffinches and blackbirds, indicating that the temporal variation in parasite burden was similar in the bird species examined. A small but significant decline in larval tick burdens was detected over the study period, possibly as a consequence of local deer control measures and/or climatic changes. Although temporal variation in tick burdens was similar between chaffinches and blackbirds, sources of heterogeneity within species were different. Sex and age had a significant effect on parasite burden in chaffinches but not in blackbirds, possibly due to differences in behaviour and immune function between these species. Our results suggest that these differences may need to be taken into account when using passerine bird species as sentinels to monitor long-term changes in tick populations.

5:15 PM - 5:30 PM (15 mins)

The effect of climate perturbations on parasite life-history variables *

Emma Gillingham[1,2], Cable J.[1], Rizzoli A.P.[2], Perkins S.E.[1,2]

[1.] Sir Martin Evans Building, School of Biosciences, Museum Avenue, Cardiff University, Cardiff, UK. CF10 3AX; [2.] Fondazione Edmund Mach, via E. Mach 1, San Michele all'Adige (TN), 38010, Italy

Parasite life-history traits are phenotypically plastic and sensitive to changes in climate, potentially affecting parasite transmission. Qualitative literature reviews provide ambiguous results, with changes in traits proving to be particular to specific host-parasite associations. Using previously published literature, we quantified the effects of 'climate change' on three parasite life-history traits: abundance, fecundity and development time. A meta-analysis of 443 data points from 36 publications found temperature increases from 'ambient' had no effect on parasite abundance, but significantly reduced fecundity and accelerated development time. Conversely, when temperatures reduced, parasite abundance increased, yet fecundity and development time were not affected. For ecto- and endoparasites, development times were significantly faster when temperatures increased for both parasite types. When temperatures decreased, however, development times of endoparasites were slower, but those of ectoparasites were significantly faster at <10.1°C below ambient temperature. Host type was important when temperatures increased; resulting in accelerated development time of parasites infecting ectothermic and endothermic hosts, yet temperature decreases did not affect development times of either parasite type. At a broad taxa level, reduced temperatures slowed development time of parasites infecting fish, but parasites of mammals and reptiles were not significantly affected. Humidity perturbations were investigated with regard to parasite development time only, due to a low sample size. Humidity increases did not significantly affect parasite development times, yet decreases resulted in faster development. Overall, our results suggest that for any given parasite exposed to raised temperatures, abundance is unlikely to change, fecundity will be reduced but development will be faster.

Session F6 (RCJ) - Emerging Vaccines

Chair: Al Nisbet, Moredun Research Institute

4:00 PM - 4:30 PM (30 mins)

Development of a subunit nematode vaccine: antigen discovery, antigen generation and adjuvant development

Tom McNeilly, Collette Britton, David Frew, Cassandra Longhi, Jacqueline B. Matthews & Alasdair J Nisbet
Moredun Research Institute

Infections with parasitic nematodes have a significant impact on animal health. Despite considerable efforts, the development of vaccines to control these parasites has been problematic; while some success has been achieved using native parasite antigens, which are often impractical for large-scale vaccination programmes, immunization with recombinant versions of these antigens has largely been unsuccessful. This may be due to a number of factors, including the use of single rather than combinations of antigens, the generation of recombinant antigens which differ significantly from the native versions, or the generation of suboptimal immune responses. This talk will summarise current progress towards the development of an efficacious recombinant subunit vaccine against *Teladorsagia circumcincta*, a pathogenic nematode affecting small ruminants. Our recent work has shown that immunization of sheep with a panel of eight recombinant antigens generated in either *Escherichia coli* or *Pichia pastoris* and selected as either targets of mucosal IgA responses in immune sheep, a known correlate of protection for this parasite, or for putative immunomodulatory function, results in significant reductions in mean egg output (59-70% reduction) and adult worm burdens (56-75% reduction). Current work is now focused on optimising this protective effect by (i) identification of novel vaccine targets via in vitro screening of parasite antigens for immunomodulatory function or by identifying proteins which are preferentially expressed by mucosal stages of the parasite; (ii) evaluation of nematode-based protein expression systems for the generation of vaccine antigens; and (iii) development of systemically-delivered adjuvants which induce enhanced mucosal antibody responses.

4:30 PM - 5:00 PM (30 mins)

Fasciola hepatica cathepsin L vaccines: we're not there…yet!

John P. Dalton[1]*, Grace Mulcahy[2], Jose Perez[3] & Álvaro Martínez-Moreno[3]

FP7 PARAVAC Consortium: [1]School of Biological Sciences & Institute for Global Food Security (IGFS), Queen's University Belfast, Northern Ireland; [2]Veterinary Medicine, University College Dublin, Dublin, Republic of Ireland; [3]School of Veterinary Medicine, University of Cordoba, Córdoba, Spain

It has been 20 years since we first patented cathepsin L proteinases as vaccines to protect ruminants from infection with the helminth Fasciola hepatica. At that time we showed that native molecules formulated in FCA/FIA induced high levels or protection against an experimental challenge infection and also elicited high anti-fecundity effects. Significant progress been made in the recombinant production of the parasite cathepsin L, and in understanding their biochemistry and how this vaccine may work. In addition, we have demonstrated the potential for a recombinant vaccine to work in field conditions. However, we have not quite reached the levels of protection that our first trials had anticipated…not yet! In this talk I will discuss the history and progress of the vaccine development using cathepsin L proteinases, experiments underway as part of the FP7-funed PARAVAC Consortium, and include discussion of other potential vaccine candidates. As well, I will mentioned the hurdles that we need to overcome to end up with a vaccine that fits the 'product profile' required for marketing and commercialization.

5:00 PM - 5:15 PM (15 mins)

Population, genetic and antigenic diversity of *Eimeria*: prospects for novel vaccines

Damer Blake[1], Emily L. Clark[1,2], Fiona M. Tomley[1] and the Eimeria CIDLID Consortium
[1.]Pathology and Pathogen Biology, Royal Veterinary College, Hawkshead Lane, North Mymms, AL9 7TA, UK. [2.]Roslin Institute, Easter Bush, Midlothian, EH25 9RG, UK

Eimeria species parasites cause the disease coccidiosis, most notably in chickens where the global cost is thought to exceed US$3 billion every year. Every one of the ~60 billion chickens produced per annum is likely to be exposed to these

parasites and control is essential. Most farmers rely on chemoprophylaxis, although drug resistance develops rapidly and is now widespread. Live parasite vaccines are available, but relative cost and production capacity limit uptake to the minority egg layer and breeder sectors. In recent years interest in developing cost effective vaccines applicable for use with broilers (birds reared for meat) have been rewarded by the identification of a panel of antigens as candidates for recombinant or subunit vaccines. Translation of these vaccines to the field will depend in part on parasite population structure and the extent of pre-existing antigenic diversity, influencing opportunities for vaccine breakthrough and dissemination of resistant genotypes. For *Eimeria* these variables remain almost completely unknown. In response to this knowledge deficit a panel of recent *Eimeria* field samples has been assembled, representing 17 countries from Asia, Africa, North America, South America, Europe and Australia. Molecular species identification has revealed the occurrence of novel operational taxonomic unit genotypes across much of the Southern, but not the Northern hemisphere. Sequenom medium-throughput genotyping has been used to define population structure for *Eimeria tenella*. Targeted sequencing and in vivo vaccination/challenge has revealed limited but immunologically relevant allelic diversity for the vaccine candidates apical membrane antigen 1 (AMA1) and immune mapped protein 1 (IMP1).

5:15 PM - 5:30 PM (15 mins)

Characterising immune responses in UK dairy cattle naturally exposed to *Fasciola hepatica*

John Graham-Brown[1], Aras Kadioglu[2], Matthew Baylis[3], Diana J.L. Williams[1]

[1.] Veterinary Parasitology, Institute of Infection and Global Health/School of Veterinary Science, University of Liverpool, Liverpool Science Park iC2, 146 Brownlow Hill, Liverpool L3 5RF [2.] Department of Clinical Infection, Microbiology and Immunology, Institute of Infection and Global Health, The Ronald Ross Building, 8 West Derby Street, Liverpool L69 7BE. [3.] Department of Epidemiology & Population Health, Institute of Infection and Global Health, University of Liverpool, Leahurst Campus, Chester High Road, Neston, Cheshire CH64 7TE.

Fasciola hepatica is a common parasite of livestock in the UK causing disease and economic losses. Vaccine trials are underway, however little is known about host-parasite interactions in a natural setting, and consequently how effective vaccines would be in the field. We recruited three cohorts of dairy heifers (n=42) previously unexposed to *F. hepatica*, and sampled them monthly over a grazing season (May to October). Disease status was determined via antibody ELISA and faecal egg counts. Peripheral blood mononuclear cell (PBMC) phenotype and parasite-specific cytokine responses were measured to characterise the immune response. Analysis comparing seropositive and seronegative animals showed an elevation in circulating lymphocytes ($p<0.0001$), with increased numbers of both CD4+ ($p<0.0001$) and CD8+ ($p<0.05$) T-cells and an overall increase in CD4:CD8 ratio in seropositive vs. seronegative samples ($p<0.005$). Additionally, parasite-specific IL-4 production showed an increase ($p<0.01$), whilst no such difference was found overall with IFN-gamma production. There was a significant increase in the eosinophil count in seropositive animals ($p<0.001$), whilst neutrophils and CD14+ monocytes decreased in peripheral circulation ($p<0.01$ & $p<0.05$ respectively). The increase in parasite-specific IL-4 production and increased CD4:CD8 ratio suggest a polarised type-2 immune response in dairy calves naturally exposed to infection. Liver fluke vaccines currently under development rely on induction of a type-1 immune response. The impact of natural infection on vaccine-induced responses requires further analysis.

Day 3 - 09/04/2014

Session A7 (WRA) - Kinetoplastida - Respond to Abstracts

Chair: Mark Field, University of Dundee

11:00 AM - 11:15 AM (15 mins)

Novel components of the mitochondrial segregation machinery and their hierarchy uncovered in *Trypanosoma brucei*

Torsten Ochsenreiter[1], Nicholas Doiron[1], Roman Trikin[1], Felix Schnarwiler[3], Achim Schnaufer[4], Benoit Zuber[2], Andre Schneider[3]

[1.]Institute of Cell Biology, [2.]Institute of Anatomy, [3.]Department of Chemistry and Biochemistry, University of Bern, Baltzerstrasse 4, 3012 Bern, Switzerland; [4.]Institute of Immunology & Infection Research and Centre for Immunity, Infection and Evolution, University of Edinburgh, Edinburgh EH9 3JT, UK

Trypanosomes contain a single mitochondrion with a single unit genome (kDNA) that is physically linked to the single flagellum of the cell. Based on electron microscopy studies this structure has been termed the tripartite attachment complex (TAC). The TAC consists of unilateral filaments that link the kDNA to a differentiated region of the mitochondrial inner membranes. A second set of filaments then associates the outer mitochondrial membrane to the basal body. Until now three components of the TAC have been described however their interactions and functions remain elusive. Through a combination of proteomics, transcriptomics and bioinformatics we have identified new components of the TAC. We demonstrate the precise localization of the components throughout the cell cycle and can show that loss of the components leads to impaired segregation but not replication of the kDNA. Furthermore for the first time we decipher the hierarchy of the currently known components and present a model of the TAC assembly.

11:15 AM - 11:30 AM (15 mins)

Discovering single nucleotide polymorphisms and structural variations in homogeneous and heterogeneous populations of trypanosomatids

Hideo Imamura[1], Tim Downing[2], An Mannaert[1], Eliane Tihon[1], Jan Van Den Abbeele[1], Jorge Arevalo[2], James Cotton[3], Shyam Sundar[4], Suman Rijal[5], Matt Berriman[3], Jean-Claude Dujardin[1]

[1]Institute of Tropical Medicine, Antwerpen, Belgium; [2]Instituto de Medicina Tropical Alexander von Humboldt, Lima, Peru; [3]College of Science, National University of Ireland, Galway, Ireland; [4]Wellcome Trust Sanger Institute, Hinxton, UK; [5]Banaras Hindu University, Varanasi, India; 6B.P. Koirala Institute of Health Sciences, Dharan, Nepal.

COCALL (COnsensus of SNP CALL Among Populations) was developed and applied to identify single nucleotide polymorphisms among recent and homogenous populations as well as more ancient and highly heterogeneous populations. The consensus method overcomes the lack of verified SNP data bases and makes use of existing SNP callers such as pileup, mpileup, Freebayes, GATK and CORTEX to identify reliable SNPs. To demonstrate its suitability for SNP analysis, we used as models (i) 191 *Leishmania donovani* isolates from the Indian subcontinent (2,418 SNPs), (ii) 54 *L. brasiliensis* isolates from Peru and Bolivia (361,031 SNPs) and (iii) 39 *Trypanosoma congolense* isolates from Sub-Saharan Africa (651,541 SNPs). A python visualization scheme for the detection of large structural variants was developed: it is well suited for genome analysis of *Leishmania* genomes, which have extensive aneuploidy, thus complicating depth variation analysis. We also provide an integrative genetic variant visualization scheme using circos for viewing complex genetic variants among populations. These flexible tools are well suited for analysis for genomes of trypanosomatids and protozoan parasites.

11:30 AM - 11:45 AM (15 mins)

Using NextGen data to model kDNA segregation and predict guide RNA genes in *Trypanosoma brucei* *

Sinclair Cooper, Achim Schnaufer, Nick Savill
Institute for Immunology and Infection Research, and Centre for Immunity, Infection and Evolution, University of Edinburgh, Edinburgh, United Kingdom

The mitochondrial genome of *Trypanosoma brucei* is unique: it consists of a large network of interlinked circular DNA molecules called the kinetoplast. This network is made up of two types of molecules; maxicircles and minicircles. Maxicircles are large (~23kb) molecules that code for genes of the respiratory chain; many of the transcripts generated from maxicircles require post-transcriptional modification by uridine insertion/deletion before translation can take place. This post-transcriptional modification of maxicircle transcripts is mediated by guide RNAs (gRNAs) encoded in minicircles. These are ~1kb in size and present in very high numbers (~10,000 per cell). There are an estimated 200-300 different classes of minicircles, each class encoding a different set of 3-5 gRNAs. The true complexity of minicircles is not known, but there appear to be high levels of redundancy and significant fluctuations in copy number over time. High minicircle heterogeneity is crucial for survival of *T. brucei*. How *T. brucei* maintains minicircle complexity is unclear, but it is thought that segregation of replicated minicircles into daughter cells is non-random. Next generation sequencing allows longitudinal investigation of minicircle and guide RNA copy numbers and quantitative investigation of the degree to which *T. brucei* controls kinetoplast network segregation. Here bioinformatics approaches have been used to generate a putative gRNA identification and quantification pipeline. This pipeline has been used to exploit existing and new short read data sets from *T. brucei* whole genome sequencing projects to determine their complete minicircle repertoire and their expression products.

11:45 AM - 12:00 PM (15 mins)

The effect of *Leishmania major* infection on atherogenesis and cytokine patterns in resistant and susceptible mice

Marc Karam, Mirna Chahine , Amani Chahine
Faculty of Sciences, Biology Department, University of Balamand, Lebanon

The outcome of the infection *Leishmania major* depends on the type of the immune response mounted by the host. This response is either cell mediated or humoral making the host resistant (as humans and C57BL/6 mice) or susceptible (BALB/c mice) to this parasite respectively. Activation of either branch of the immune system depends on other factors such as genetic makeup of the host, the infecting dose of parasites and the cytokine milieu (during early stages of the infection). In BALB/c mice, the early production of Th2 cytokines (TGF-β, IL-4, IL-5, IL-10 and IL-13) inhibits IL-12 function and natural killer (NK) cells which are crucial for Th1 cell activation. On the other hand, L. major infection in resistant hosts results in Th1 cells activation with production of IL-2, Interferon-γ (IFN-γ) and Tumor Necrosis Factor-α (TNF-α). On the other hand, inflammation plays an important role in atherosgenesis whereby T helper cells seem to play a role in the plaque development. Th1 cells produce mainly IFN-γ, TNF-α and IL-2 which lead to the activation of macrophages, induction of endothelial dysfunction and promotion of plaque destabilization. While Th2 cells produce mainly IL-4, IL-5, IL-13 and IL-25 and seem to have an atheroprotective role because they suppress the Th1 pro-inflammatory response. We hypothesize here that Th1 and proinflammatory cytokines that confer protection against this parasite can promote atherogenesis in resistant hosts while Th2 cytokines might have protective effects against atherosclerosis while making the hosts susceptible to *L. major* infection.

12:00 PM - 12:15 PM (15 mins)

The dynamics of mitochondrial RNA binding complex in *Trypanosoma brucei* and its petite mutant under optimized immobilization conditions

Hassan Hashimi[1,2], Zhenqiu Huang[1,2], Sabine Kaltenbrunner[2], Eva Šimková[3], David Staněk[3], Julius Lukeš[1,2]
[1] Institute of Parasitology, Biology Centre, Czech Academy of Sciences, České Budějovice (Budweis), Czech Republic [2] Faculty of Sciences, University of South Bohemia, České Budějovice (Budweis), Czech Republic; [3] Institute for Molecular Genetics, Czech Academy of Sciences, Prague, Czech Republic

There is a variety of complex metabolic processes ongoing simultaneously in the single, large mitochondrion of *Trypanosoma brucei*. Understanding the organellar environment and dynamics of its proteins requires quantitative measurement in vivo. In this study, we have validated a method for immobilizing both procyclic (PS) and bloodstream (BS) stages of *Trypanosoma brucei brucei* with a high level of cell viability over hours and verified its suitability for undertaking fluorescence recovery after photobleaching (FRAP), using mitochondrial-targeted yellow fluorescent protein

(YFP). Mobility of this protein appears to be hindered in the PS as compared to the BS, possibly due to cristae that extend into the mitochondrial matrix only in the former stage. Next, we used this method for comparative analysis of the motility of mitochondrial RNA binding protein 1 (MRP1) in the BS and in *T. brucei evansi*, which is its petite mutant lacking organelle-encoded nucleic acids. FRAP measurement of YFP-tagged MRP1 revealed its different dynamics in both cell lines, illuminating from a new perspective how the absence or presence of RNA affects proteins involved in mitochondrial RNA metabolism. This work represents the first attempt to examine this process in live trypanosomes.

12:15 PM - 12:30 PM (15 mins)

What do kinetoplastids need a kinetoplast for? Life cycle progression of *Trypanosoma brucei* in the presence and absence of mitochondrial DNA *

Caroline Dewar, Achim Schnaufer

Centre for Immunity, Infection and Evolution, Institute of Immunology and Infection Research, King's Buildings, University of Edinburgh, Edinburgh, EH9 3JT, UK

We recently showed that specific mutations in a nuclearly encoded subunit of the mitochondrial FoF1-ATPase negate the requirement for mitochondrial DNA (the kinetoplast or kDNA) in long slender bloodstream form (LS) *Trypanosoma brucei* (Gould *et al.,* 2012). We are now investigating two questions: (1) Are kDNA and a functional FoF1-ATPase required for life cycle progression? (2) What is the molecular mechanism of compensation for kDNA loss? The fundamental differences between energy metabolism in procyclic insect form (PF) and LS *T. brucei* involve a switch in the directionality of the FoF1-ATPase. In PF, oxidative phosphorylation requires the enzyme to generate ATP. In LS, the enzyme uses ATP from glycolysis to drive proton pumping to maintain the essential mitochondrial membrane potential. Fo-ATPase subunit 6 is critical for proton translocation in either direction and is kDNA-encoded. We have shown that a mutated F1-γ subunit allows LS viability in the absence of kDNA. We have generated pleomorphic (i.e. differentiation-competent) *T. brucei* with and without kDNA by expressing mutant g in strain AnTat 1.1. Differentiation studies demonstrate that these cells do not require kDNA for formation of transmissible stumpy forms. However, viability of stumpy forms is impaired in kDNA0 cells, suggesting a critical role for a kDNA-encoded product in these cells. Likewise, we find that kDNA is indispensable for progression to the PF form. Further, we have generated LS *T. brucei* expressing affinity-tagged ATPase subunits and mutant γ and will discuss how γ mutation and kDNA loss, respectively, affect structure/function of the FoF1-ATPase.

Session B7 (WRR) - Apicomplexa - Immunology

Chair: Julius Hafalla, London School of Hygiene and Tropical Medicinea Lawniczak, Imperial College London

11:00 AM - 11:30 AM (30 mins)
Immune responses and virulence in *Plasmodium chabaudi* infection
Jean Langhorne
MRC National Institute for Medical Research, London, UK

Immune responses to the blood stages of *Plasmodium* have been well defined in mouse models, particularly *Plasmodium chabaudi chabaudi* (AS). There is a requirement for innate responses, CD4 T cells and antibody as effector mechanisms. However these responses can also play a role in the pathology, and therefore need to be tightly controlled. Much of our knowledge about the responses and their regulation comes from experiments initiated by infections initiated by direct blood challenge. To make the model as relevant as possible to human malaria, we have investigated the host response in blood-stage infections using mosquito-transmitted infections. We demonstrate that mosquito transmission (MT) attenuates the virulence and parasitaemia of blood-stage infections, which is dependent on the presence of an intact host immune system. Despite attenuation, many of the well-known characteristics of a *P. chabaudi* infection are still observed; chronic infection, differently virulent parasite strains, and susceptible and resistant mouse strains. The attenuated MT infections are accompanied by an up-regulation of transcription of the cir multigene family, which encodes a family of variant proteins. We suggest that CIR proteins play a role in interacting with the host immune system, and thus determining the virulence of blood-stage *P. chabaudi* infections

11:30 AM - 11:45 AM (15 mins)
Localising Selection from Resequencing Data: Linking Genes to Phenotypes in Malaria Parasites
Chris Illingworth[1], Andrej Fischer[2], Megumi Inoue[3,4], Hussein Abkallo[4], Axel Martinelli[2], Paul Hunt[5], Ho Y. Shwen[6], Arnab Pain[6], Richard Culleton[4], Ville Mustonen[2]
[1]Department of Genetics, University of Cambridge, UK; [2]Wellcome Trust Sanger Institute, Hinxton, Cambridge, UK; [3]Department of Protozoology, Institute of Tropical Medicine, Nagasaki University, Nagasaki, Japan; [4]Malaria Unit, Institute of Tropical Medicine, Nagasaki University, Nagasaki, Japan; [5]Institute of Immunology and Infection Research, University of Edinburgh, Edinburgh, UK [6]Computational Bioscience Research Center, Chemical Life Sciences and Engineering Division, King Abdullah University of Science and Technology, Thuwal, Kingdom of Saudi Arabia.

Genetic crosses are a valuable resource for identifying genetic causes for specific phenotypes. Exposing a cross population to different selection pressures leads to characteristic changes in allele frequencies, which can be analysed to identify segregating sites of functional importance. However, given experimental noise from a variety of sources, identifying the precise locations of alleles under selection pressure presents a statistical challenge. We here present a novel approach to localising alleles under selection from genome sequence data collected from a genetic cross between strains of *Plasmodium yoelii* grown in naive and vaccinated mice. Our method filters potentially mis-mapped reads, shortlists genomic regions exhibiting non-neutral behaviour, and identifies selected alleles using a hierarchical family of multi-locus evolutionary models, which account for variance in the local recombination rate. We combine data from different experimental replicates to generate confidence intervals for the location of alleles under selection. Applying our method, we identify candidate genes in the parasite for increased growth rate and for evasion of the host immune response.

11:45 AM - 12:00 PM (15 mins)

A comparative transcriptomic and proteomic investigation of host cell responses during *Toxoplasma gondii* and *Neospora caninum* invasion of human astrocytes. *

Sarah Altwaim, Xia D and Wastling J

Department of Infection Biology, Institute of Infection and Global Health, University of Liverpool. Liverpool Science Park IC2, 146 Brownlow Hill, L3 5RF, Liverpool

Toxoplasma gondii and *Neospora caninum* are intracellular protozoan parasites from the phylum Apicomplexa. Both parasites share many morphological and genetic features, but have diverse host preferences. While *T. gondii* can infect any warm-blooded animal including humans, *N. caninum* does not and is widely spread through vertical transmission in cattle. In immunosuppressed individuals, the chronic stage of *T. gondii* can cause Toxoplasma encephalitis, whereas neosporosis results in neuromuscular disease in dogs and abortion in cattle. The basis of host preference in these two parasites is unknown, but could be due to differences in tropism to specific host cells. To test this hypothesis, we investigated the differential expression of host-cell and parasite genes/proteins during the invasion of human astrocyte (HA) cells by both *Toxoplasma* and *Neospora*. Parasites and HAs were cultured in vitro; and infected with type III *T. gondii* and *N. caninum* Liverpool isolate strain. Samples were collected at early stages of infection for both transcriptomic and label-free proteomic analysis. A number of differences in host cell responses were noted between *Toxoplasma* and *Neospora* infection. Among these differences in expression were metabolic pathways involved in Extracellular Matrix receptor interaction found in *Toxoplasma* infected cells, whereas Fc gamma R-mediated phagocytosis was found in *Neospora* infected cell. Transcriptomics data from *T. gondii* suggests the up-regulation of various suppressive factors that prevent activation of the host response. These data suggest that the two parasites stimulate some fundamentally different host cell responses. Further investigation of host secretome during infection will be explored.

12:00 PM - 12:15 PM (15 mins)

Parasitology and inflammation in kidneys and lungs in a murine model of co-infection *Plasmodium*/filarial nematode *

Gregory Karadjian[1], D. Berrebi[2], I. Landau[1], O. Bain[1], C. Mantin[1]

[1]*Museum National d'Histoire Naturelle, UMR 7245, 61 rue Buffon, CP52, 75005 Paris.* [2]*Service d'Anatomie et de Cytologie Pathologique, Paris, Hôpital Robert Debré, Assistance Publique-Hôpitaux de Paris France, and EA3102, Université Paris 7, France.*

Malaria and helminths infections, including filarial nematodes, are two of the most prevalent parasitic diseases in the tropics and co-infection is clearly the norm rather than the exception. However the interactions between the different parasites in the same host remain complex and poorly understood. Despite increasing activity in recent years, studies comparing co- and mono-infections are very much in their infancy and results are potentially contradictory. In this study, BALB/c mice were concomitantly infected with the filarial nematode *Litomosoides sigmodontis* and *Plasmodium yoelii yoelii* 17 XNL or *Plasmodium chabaudi chabaudi* 864VD. During the course of infection, a parasitological study was performed as well as an analysis of the inflammation including anatomo-pathological observations in kidneys and lungs, and measurement of early inflammatory series cytokines. Whereas the filarial recovery rate was strongly decreased in mice co-infected with both *Plasmodium* species, the peak of parasitaemia of *P. yoelii* was decreased in co-infected mice but not the peak of *P. chabaudi*. At day 4 and day 7 post-inoculation, levels of IFN-γ and TNF-α were increased in mice infected with *P. yoelii* and co-infected. *L. sigmodontis* can reverse lesions in the kidneys due to the presence of both *Plasmodium* species but does not modify the course of pulmonary lesions. At day 7 only, an original transitory observation in the lungs of mice infected by *L. sigmondis*, or co-infected, is the presence of granulomas, containing neutrophils, T cells and macrophages.

12:15 PM - 12:30 PM (15 mins)

No Evidence that Knops Blood Group Polymorphisms Affect Complement Receptor 1 Clustering on Erythrocytes *

Olivia Swann[1], Opi DH[2], Harrison EM[3], Nyatichi E[2], Tendwa M[2], Macharia A[2], Uyoga S[2], Williams T[2,3], and Rowe JA[1]

[1.]Centre for Immunity, Infection and Evolution, Institute of Immunology and Infection Research, School of Biological Sciences, University of Edinburgh, Edinburgh, UK; [2.]Wellcome Trust Research Laboratories/Kenya Medical Research Institute, Centre for Geographic Medicine Research, Kilifi, Kenya; [3.]Centre for Inflammation Research, Edinburgh, UK d Department of medicine, Imperial College, London, UK

Background Complement receptor 1 (CR1) on erythrocytes is a ligand for *Plasmodium falciparum* erythrocyte receptor 1 (PfEMP1). CR1 clusters on the erythrocyte membrane on binding a ligand, increasing avidity. The combined genotype of Swain-Langley 2 (Sl 2/2) and McCoy a (McC a/a) CR1 polymorphisms has been associated with protection against cerebral malaria (OR 0.51, p=0.012) and mortality (OR 0.33, p=0.016) when compared to the Sl 1/1 McC a/a genotype in Kenyan children. The addition of McC b alleles negated this. We postulated these genotypes affected CR1 clustering, influencing its ability to bind and clear opsonised immune complexes. Methods We developed an assay to quantify number and volume of CR1 clusters on an individual's erythrocytes using immunofluorescence, confocal microscopy and image analysis software. The assay was performed on fresh venous blood samples from children in the Kilifi district of Kenya. 29 children had Sl 1/1 McC a/a, 88 had Sl 2/2 McC a/a and 8 had Sl 2/2 McC b/b genotype. Mean CR1 copy number per erythrocyte, alpha-thalassaemia and sickle-cell genotypes were determined. Hierarchical mixed effect Poisson and linear regression models were used. Results A strong linear correlation was observed between CR1 copy number and median number of CR1 clusters per erythrocyte and median cluster volume. After adjustment for CR1 copy number, alpha-thalassaemia and sickle cell genotype, Sl/McC genotype had no effect on number of CR1 clusters per erythrocyte or cluster volume. Conclusion The Sl 2/2 McC a/a genotype does not appear to be the protecting against cerebral malaria through altered CR1 clustering.

Session C7 (WRL) - Vectors and Parasites in the Middle East - Vectors and Parasites in the Middle East II

Chair: Mahmoud N. Abo- Shehada, London School of Hygiene and Tropical Medicine

11:00 AM - 11:30 AM (30 mins)

Sarcosporidia and Sarcosporidiosis (Apicomplexa: Coccidia) infecting reptiles in Egypt and Saudia Arabia

Fathy Abdel-Ghaffar[1], Abdel-Rahman Bashtar[1], Kareem Morsey[1], Rewaida Abdel-Gaber[1], Saleh Al-Quraidhy[3], Heinz Melhorn[2]

[1] Zoology Department, Faculty of Science, Cairo University, Cairo, Egypt; [2]Parasitology Institute, Düsseldorf University, Düsseldorf, Germany Zoology Dept., [3]College of Science, King Suad University,Riyadh, Suadia Arabia

Parasitological studies carried out on reptiles are of a special interest because they are a very ancient group in the evolutionary scale and their parasites seem to become stabilized all over the world. Among their parasites, the genus *Sarcocystis* is the most widely distributed group. Even moderate infection of these organisms may cause the reptiles to stop feeding and may die. During the period from January 2012 to October 2013, 430 Acanthodactylus lizards and 350 Chalcides skinks from different localities were examined for micro- and macroscopic Sarcosporidia. Two forms of Sarcocystis species were recorded, macroscopic cysts which were demonstrated in two species of Acanthodactylus (*A. boskianus* and *A. paradalis*) with a percentage of 11.6 and microscopic cysts in two species of Chalcides (*C. ocellatus* and *C. chalcides*) with a percentage of 21.4. Light microscopic study revealed that the macroscopic cysts measured 0.95 x 0.12 mm and bounded by a thick primary striated cyst wall measured 3.9µm. The cyst wall was characterized by the presence of finger like stalkless protrusions, measured 2.65 x 0.84µm and with granulated ground substance while microscopic cysts infecting *Chalcides* spp. measured 77.5 x 562.5 µm and bounded by a thick primary striated cyst wall measured 5.8 µm. The cyst wall was characterized by the presence of long leaf like protrusions measured 2.4 µm. Ultrastructurally, the interior of the cysts for both forms was occupied by many banana shaped merozoites, each measured 4.8 x 1.85 µm and 4.5 x 1.35 µm respectively. Metrocytes were located at the periphery of the cyst and measured 3.95 x 3.6 µm for macroscopic cysts and 2.75 x 5.5 µm for microscopic cysts. Transmission experiments were done from the infected Acanthodactylus and skink Chalcides spp. to the snake *Spalerosophis diadema*. In *Acanthodactylus* spp., shedding of sporulated oocysts and sporocysts was at the 16th day p.i. till the 35th day. Gamogony takes place within the intestinal epithelial cells of the final host (snake) through the first four days p.i. while sporogony occurred in the lamina propria of the small intestine of the inoculated snakes. The sporulated oocysts measured 10.9 x 16.3 µm and were seen beginning at the 14th day p.i. The skin of the infected skinks appeared pale for the first time, while the infected snakes with Sarcocystis showed loss of appetite, weakness, weight loss and acute anemia. These pathogenic effects progressed in severity until some infected snakes were died.

11:30 AM - 11:45 AM (15 mins)

Parasitology research in the Middle East and North Africa 1950-2013

Mahmoud Abo-Shehada

London School of Hygiene & Tropical Medicine

Systematic search for parasitology research output produced between 1950 and 2013, and 2004 to 2013 by the 19 countries of the Middle East and North Africa (MENA) was carried out using the Web of Science of Thomson Scientific's Essential Science Indicators database. The parasitology research in MENA countries was ranked based on the number of articles, average citation per article and Hirsch-Index (H-Index). During the 63 years, the 19 countries produced 13746 parasitological articles with 82% by Egypt, Israel, Iran and Saudi Arabia and six were in the 75% quartiles of H-Index: Israel (79), Egypt (41), Iran (38), Saudi Arabia (29), Lebanon (27) and Tunisia (27). In the last ten years 38.2% of articles were produced with Iran and Tunisia being most improved and Iran enhancing her rank from third to second in the H-Index. When normalized to population and growth domestic product per capita, the output of high income countries lagged behind middle income countries. The main researched parasite groups were Protozoa, Trematoda, Nematoda, Cestoda, Acari and diptera, in decreasing order. Articles on endo-parasites were approximately double those of ecto-parasites. More than half of MENA endo-parasite research was on *Leishmania* spp., *Schistosoma* spp., *Echinococcus* sp., *Toxoplasma gondii* and *Plasmodium* spp. During the last ten years, cestoda articles increased from 7% to 11% at the expense of trematoda articles. In MENA, parasitology research strengths and needs were identified.

11:45 AM - 12:00 PM (15 mins)

***Bacillus thuringiensis* Induces Cellular Stress in the Mosquito Vector, *Culex pipiens*, Prior to Death**

Ashraf Ahmed
King Saud University, College of Science, Zoology Department, Riyadh, Saudi Arabia

This study was conducted to investigate the oxidative stress and apoptotic signs detectable by flow cytometry as proposed pathogenicity mechanisms for the mosquitocidal bacterium *Bacillus thuringiensis* (Bt) in the mosquito vector, *Culex pipiens*. Obtained data showed elevation in the levels of the oxidative stress biomarkers, the lipid peroxidation and protein oxidation, upon Bt-infection. Larvae showed significant higher levels of both lipid peroxidation and protein oxidation at 12 and 24h post-infection compared to control ones. In addition, Bt-inoculated adult mosquitoes also showed significant higher lipid peroxidation at 12 and 24h post-inoculation compared to control ones. These signs of oxidative stress were more pronounced in bacterial infected larvae than in bacterial inoculated adult mosquitoes. Finally, Bt-infected larvae showed significant higher percentages of cellular apoptosis at 12 an 24h post-infection compared to control ones. These data may indicate that Bt infection induced oxidative stress and apoptosis proceeding cellular damage, and thus, may be suggested as important pathogenicity mechanisms of Bt in its mosquito host. And hence, these data may participate in improving our understanding of the mosquito-Bt interaction scenario, which may help improving the biocontrol measurements against mosquito vectors.

12:00 PM - 12:15 PM (15 mins)

Insecticidal Activity of Newly Isolated Actinomycete Strains from the Desert Habitats of Saudi Arabia Against *Culex pipiens*

Wael Hozzein, Fahd A. Al-Mekhlafi, Mohamed A.M. Wadaan
College of Science, King Saud University, P.O. Box 2455 Riyadh 11451, Kingdom of Saudi Arabia.

Insect vectors of diseases were and still one of the most pressing problems which affect human health and the environment. Therefore, it is still a big demand to try to find new environmentally safe biocontrol agents to manage insect vectors. The present study was carried out to evaluate the insecticidal activity of some new actinomycete strains isolated from the desert habitats in the Eastern Province of Saudi Arabia. Based on a preliminary test and the antimicrobial activities, four strains were chosen for the larvicidal activity of their crude extracts against the 2nd instar larvae of *Culex pipiens* reared in the laboratory. The four strains were identified by studying their phenotypic characteristics and the phylogenetic analysis of the 16S rRNA gene sequence. The strains were identified as *Actinomadura* sp. GD15, *Nonomuraea* sp. GB14, *Streptomyces* sp. GB12 and *Streptomyces* sp. GO23. At 200 ppm, 100% mortality in larvae was obtained with the four extracts after 24 h of exposure and the highest larval mortality was found with the extract of *Streptomyces* sp. GB12 (LC50=56.74±0.66 ppm). The present investigation clearly reveals the insecticidal potentials of the selected actinomycete strains and one of them can be identified as a potential biocide producer but further studies are required to exploit these results for human welfare.

12:15 PM - 12:30 PM (15 mins)

Dynamic trends in intestinal parasitic infections among recently arrived immigrant workers, settled immigrants and long-term residents in Qatar.

Jerzy Behnke, Marwan A. Abu-Madi, Sanjay H. Doiphode
Qatar University, Hamad Medical Corporation, University of Nottingham

The expanding economy of Qatar has attracted immigrants, often from countries with poor socio-economic standards. Many arrive with intestinal parasitic infections. The prevalence of infection among newly arrived immigrants was 26.5% compared to 16.5% among residents, the biggest drop being among helminths (20% to 9.2%). Among newly arrived workers prevalence varied significantly by region of origin, and hookworm infections in particular were frequent among the Nepalese but in contrast to all other regions, did not fall markedly after acquisition of residency status. Protozoan infections changed little overall, because some species increased while others declined: e.g. a significant increase in prevalence of *Blastocystis hominis* among residents compared with immigrants and a concomitant declined in both *Giardia duodenalis* and *Entamoebae histolytica/dispar*. We analyzed also another data-set of 18,563 hospital records of subjects from 57 countries who between 2005 and 2011 sought medical assistance. Overall 8.6% were infected with one or more species, but in the last three years there were falling trends of prevalence providing some optimism that parasitic infections among the resident immigrants have begun to decline. We identified also geographic regions from which resident workers still maintain a relatively high prevalence of helminth infections. Workers from Nepal were the most likely to carry hookworm infections (19.7%) followed by other W. Asian nationals including in descending order those from Bangladesh (8.3%), Sri Lanka (6.8%) and India (4.4%). Our results have clearly identified high risk groups among the many foreign immigrant workers and have clear implications for the health authorities.

Session D7 (RCA) - Helminths - Helminths Chemotherapy & Drug Targets

Chair: Russell Stothard, Liverpool School of Tropical Medicine

11:00 AM - 11:30 AM (30 mins)
Schistosomiasis and Praziquantel - past and future of a gold standard chemotherapy
Jutta Reinhard-Rupp
Merck Serono S.A.

The helminthic disease schistosomiasis is considered as one of the most neglected diseases that impacts human health and history since ancient times. Today more than 240 Million people, mainly in sub-Saharan Africa, are infected and an estimated number of more than 200,000 people die every year of this disease that could have been eliminated for decades. Treatment is possible and the gold standard today is praziquantel which becomes more and more accessible and affordable in low income countries. Still, Praziquantel is the only low-cost treatment available and no drug pipeline has been built in the past. What do we know about the current mechanism of action for praziquantel, what would be the alternatives and why is this chronic disease still a high burden for so many developing countries? Questions will be analyzed and points of view will be provided during the session.

11:30 AM - 11:45 AM (15 mins)
Oxantel pamoate against *Trichuris trichiura* infections *
Benjamin Speich[1], Shaali M. Ame[2], Said M. Ali[2], Rainer Alles[3], Jörg Huwyler[3], Jan Hattendorf[4], Jürg Utzinger[4], Marco Albonico[5], Jennifer Keiser[1]
[1.] Department of Medical Parasitology and Infection Biology, Swiss Tropical and Public Health Institute, and University of Basel, Basel, Switzerland; [2.] Laboratory Division, Public Health Laboratory (Pemba)–Ivo de Carneri, Chake Chake, Tanzania; [3.] Department of Pharmaceutical Sciences, Division of Pharmaceutical Technology, University of Basel, Basel, Switzerland; [4.] Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, and University of Basel, Basel, Switzerland; [5.] Ivo de Carneri Foundation, Milano, Italy

Infections with the three soil-transmitted helminths (*Ascaris lumbricoides, Trichuris trichiura* and hookworm) are widespread and often occur concomitantly. The standard treatments albendazole and mebendazole show low efficacy against *T. trichiura* infections. Oxantel pamoate has excellent trichuricidal properties, and hence might be an interesting partner drug in combination chemotherapy. In a randomized controlled, double-blind trial conducted on Pemba Island, Tanzania we randomly assigned children, 6 to 14 year of age, to receive one of four treatments:(i) oxantel pamoate (20 mg/kg) – albendazole (400 mg) combination administered on two subsequent days; (ii) single oxantel pamoate (20 mg/kg); (iii) single albendazole (400 mg); and (iv) single mebendazole (500 mg). Cure rates (CRs) and egg reduction rates (ERRs) were calculated by available case analysis. Adverse events were assessed four times after treatment. From 458 children complete data were available, of whom 450, 443 and 293 were infected with *T. trichiura*, hookworm and *A. lumbricoides*, respectively. In the treatment of *T. trichiura* infections, oxantel pamoate combined with albendazole had significantly higher CR and ERR compared to mebendazole. Single albendazole had further significantly lower CR and ERR than mebendazole. Albendazole showed high efficacy against hookworm and *A. lumbricoides*, while mebendazole had only high efficacy against A. lumbricoides. Oxantel pamoate revealed low efficacy against hookworm and *A. lumbricoides*. Most adverse events were mild without noteworthy differences observed between treatment arms. In conclusion, Oxantel pamoate-albendazole shows higher CR and ERR against the most common soil-transmitted helminths; hence this combination could have a significant impact within global soil-transmitted helminthiasis control programs.

11:45 AM - 12:00 PM (15 mins)
Molecular diagnosis of anthelmintic resistance in parasitic nematodes
Roger Prichard
McGill University, Institute of Parasitology, Montreal, Canada

Anthelmintic resistance has become a serious problem for the control of parasitic nematodes in livestock. There is also recent evidence that resistance is developing in nematode parasites of companion animals, such as *Dirofilaria immitis* and in the human parasites, *Onchocerca volvulus* and *Trichuris trichiura*. Conventional methods for monitoring for anthelmintic resistance, such as faecal egg count reduction tests are insensitive, expensive and slow and often only used

in research settings. It has been known for some time that mutations in the β-tubulin gene can confer benzimidazole resistance. Genetic analyses have been applied to sheep farms to detect benzimidazole resistance. Recently, we have discovered mutations in the dyf7 gene confer macrocyclic lactone resistance in *Haemoinchus contortus* and *Caenorhabditis elegans*. While the deletion of a 63 bp indel in the acr8 gene causes truncation of the Acr8 subunit of the levamisole receptor in *H. contortus* and levamisole resistance. This new information allows us to apply genetic tests to detect resistance to all of the common broad spectrum anthelmintics used to control parasites in livestock. The availability of new genetic methods for monitoring anthelmintic resistance offers considerable advantages over conventional in vivo and in vitro methods for assessing anthelmintic resistance.

12:00 PM - 12:15 PM (15 mins)

A•WOL macrofilaricidal drug discovery and development - optimisation of anti-Wolbachia efficacy

Louise Ford[1], Gemma L. Nixon[1], Kelly L. Johnston[1], Joseph D. Turner[1], Neil G. Berry[2], Paul M. O'Neill[2], Stephen A. Ward[1] and Mark J. Taylor[1]

[1]*Parasitology Department, Liverpool School of Tropical Medicine, Liverpool, UK;* [2]*Department of Chemistry, University of Liverpool, Liverpool, UK*

There is an urgent need to develop a novel treatment for filariasis, and targeting Wolbachia provides safe macrofilaricidal activity with superior therapeutic outcomes compared to standard anti-filarial treatments. The Anti-Wolbachia (A•WOL) Consortium has developed both in vitro and in vivo assays, to screen chemical libraries for anti-Wolbachia activity. The outputs from the A•WOL program are now being pursued as part of A•WOL II Macrofilaricide Drug Discovery & Development programs. Screening of >10000 compounds from the BioFocus library and chemoinformatic analysis have generated six independent lead series chemotypes with the potential to enter a medicinal chemistry "hit-to-lead" and lead optimization program. A·WOL Drug Discovery is now progressing these lead series through a rigorous lead optimisation and candidate selection process, using iterative cycles of medicinal chemistry and biological testing in order to deliver at least one novel pre-clinical candidate and a chemically distinct back-up, aligned with our Target Product Profiles for an anti-Wolbachia macrofilaricide. In addition, ongoing screening of large diversity-based libraries (150-500k compounds) aims to provide additional, chemically diverse hits, with one-order improvement in absolute potency or significant shortening of treatment time, in order to expand the structural diversity of anti-Wolbachia chemotypes. A·WOL Drug Development is optimising regimens of anti-Wolbachia monotherapy and combination treatment of registered anti-Wolbachia and anti-filarial drugs *in vivo* using an adult *Brugia malayi* mouse model. This efficacy testing is driven by a rational PK/PD modelling approach which supports dosage regimens, in order to identify the best treatment regimens to test in field trials.

12:15 PM - 12:30 PM (15 mins)

Bridging wet and dry labs: a proof of concept for rational drug design against tropical diseases

Adriana Erica Miele[1] Boumis G., Saccoccia F., Brunori M., Bellelli A. [2]. Via A., Caroli A., Simeoni S., Tramontano A. [3]. Sayed A., Huang H.H., Williams D.L. [4]. Angelucci F.

[1] *Dept. Biochemmical Sciences & Institute Pasteur – Fondazione Cenci Bolognetti, University of Rome "Sapienza", Rome, Italy* [2]*Dept. Physics, University of Rome "Sapienza", Rome, Italy & King Abdullah University of Science and Technology, Tuwal, Kingdom of Saudi Arabia* [3]*Dept. Microbiology and Immunology, Rush University Medical Center, Chicago, USA* [4]*Dept. Healt, Life & Environmental Sciences, University of L'Aquila, Italy*

Tropical diseases represent a huge burden for human healthcare and account for the highest rate of mortality and disability of humankind. Being mostly confined in developing/emerging countries, they are set aside by both big pharma and many research-funding agencies. Here, we present a proof of concept strategy, which paves the way to the (re)discovery of FDA orphan drugs and their rational use against selected pathogen targets. It is important to use already approved drugs in order to speed up the process from in vitro experimentation to in vivo use, thus drastically cutting the costs for the introduction of new molecules in the market. We have tested this strategy on *Schistosoma mansoni*, the trematode causative agent of intestinal schistosomiasis, affecting more than 200 million people across the tropics. In particular, *in silico* experiments showed that auranofin, an organometallic compound in use against rheumatoid arthritis, can bind the parasite thioredoxin glutathione reductase (SmTGR), an essential enzyme of the ROS detoxification machinery. Based on docking results[1], we performed in vitro experiments on recombinant SmTGR, which proved successful. Inhibited SmTGR has later on been crystallized to understand the molecular mechanisms of inhibition[2] and the drug has been tested ex vivo on worm extracts and in vivo on infected mice[3]. This study indicates that compounds targeting parasite antioxidant responses could become clinically relevant drugs in the near future. [1] Caroli et al. Biochem Biophys Res Commun. 2012. 417(1):576-81. [2] Angelucci et al. J Biol Chem. 2009. 284(42):28977-85. [3] Huang et al. Curr Pharm Des. 2012. 18(24):3595-611.

Session E7 (RCU) - Ecology - Genetics, Evolution and Ecology of Host-Parasite Interaction

Chair: Ben Longdon, Cambridge University

11:00 AM - 11:30 AM (30 mins)

Global bio-insecurity breaches phylogeographic barriers leading to panzootic amphibian/chytrid parasitisms

Matthew Fisher

Imperial College London

The discovery of *Batrachochytrium dendrobatidis* (Bd) and chytridomycosis as a leading cause of amphibian mortality and population decline has clearly demonstrated the risk that chytrid fungi pose to amphibian health. However, there has always been a suspicion that Bd cannot be the only chytrid that infects amphibians. I will review recent genomics work showing that there are deep genetic differences between Bd isolates from different regions, suggesting a widespread ancient phylogeographic radiation within this species with one lineage, known as BdGPL, broadly emerging as a pathogen worldwide. Recent dieoffs of Dutch, and now Belgium, fire salamanders were not caused by Bd, leading to the recent discovery of an entirely new species of *Batrachochytrium* that parasitises amphibians. This talk will chart the potential evolutionary outputs of the 'chytrid explosion'.

11:30 AM - 11:45 AM (15 mins)

Why do flies vary in their susceptibility to infection?

Frank Jiggins

Department of Genetics, University of Cambridge

It is common to find considerable genetic variation in susceptibility to infection in natural populations. Within populations of *Drosophila*, we have found that much of this variation is caused by a small number of major-effect polymorphisms. The genes tend to highly specific in their effects, targeting specific viruses or virus genotypes. This simple genetic architecture is caused by selection by viruses in nature increasing the frequency of recently arisen major-effect alleles that increase resistance. Resistance is evolving due to 'improvements' to the flies' antiviral defences, as even the susceptible alleles of these genes confer some degree of antiviral protection. A second cause of variation in resistance are the bacterial symbionts Wolbachia, which protect many species of Drosophila against viral infection. Overall, susceptibility to viral infection is controlled by a small number of major-effect genes and symbionts.

11:45 AM - 12:00 PM (15 mins)

Covert specialism of apparently shared *Bartonella* parasites within woodland rodent communities. *

Susan Withenshaw, Godefroy Devevey, Amy Pedersen, Andy Fenton

Institute of Integrative BIology, University of Liverpool Centre for Infection, Immunity and Evolution, University of Edinburgh

Most parasites occur within multi-host communities. The magnitude and direction of parasite transmission between host species may determine the persistence of infection within particular subsets of the community; therefore understanding such transmission pathways is crucial for the effective targeting of disease control strategies. However, the structures of underlying transmission networks are often difficult to determine. One reason for this is that the same parasite species may appear to infect multiple host species, but actually circulate independently within each host species population. Such covert specialism of a parasite may therefore only be evident when infections are characterised at the DNA/RNA sequence level, revealing genetically diverse host-specific sub-types. Here, I will present evidence of covert specialism within flea-borne Bartonella parasites in natural communities of rodents. Several species of Bartonella appear to infect both wood mice (*Apodemus sylvaticus*) and bank voles (*Myodes glareolus*) within the same woodland communities in the UK, suggesting that some degree of between-species transmission is likely. However, results of large-scale transmission manipulation experiments, coupled with DNA sequence characterisation of Bartonella infections, appear to contradict some of the hypothesised transmission networks based solely on species-level parasite identification. Several potential mechanisms underlie such covert specialism within this system, including ecological barriers to between-species host contacts, and the presence of several species of flea vectors, whose specific relationships with host species and/or Bartonella types may inhibit bacterial transmission between host species.

12:00 PM - 12:15 PM (15 mins)

An experimentally evolved trypanosome and its implication for infection success and virulence in the bumblebee *Bombus terrestris* *

Monika Marxer, Paul Schmid-Hempel
Institute of Integrative Biology ETH Zurich Universitätsstrasse 16 8092 Zurich Switzerland

The outcome of host-parasite interactions is strongly affected by several factors including host and parasite genotypes. Selection on basic growth properties in parasites may therefore have far reaching consequences for infection outcome and the consequences for host fitness. It is know that strains of the trypanosome *Crithidia bombi* infecting bumblebees have widely varying growth rates when cultured in vitro. We succeeded in experimentally evolving this parasite in vitro, selecting for fast and slow growing sub-lines. This enabled us to investigate the costs, benefits and fitness trade-offs related to parasite growth rate by subsequently measuring in vivo infection profiles, host immune response, and competitive ability under co-infection. These results will help define the fitness consequences for the observed natural variation in the growth of *Crithidia bombi*, and will also inform important aspects of host-parasite evolution including the evolution of virulence and host-defense mechanisms.

Session F7 (RCJ) - Food Security

Chair: Damer Blake, Royal Veterinary College

11:00 AM - 11:30 AM (30 mins)
Parasites and Food Security: A complex relationship
Katharina Stärk
Royal Veterinary College, London

Food security is understood to include the sufficient availability of safe, wholesome and affordable food. This indicates that food safety is an integral part of food security. In addition, food security also makes reference to the nutritional aspects of food and the efficiency of food production including food prices. Because food security covers such diverse aspects of food production, there are numerous pathways by which parasites can impact on food security. Parasites can infest either humans or animals or both depending on the development cycle. For a significant number of parasites, animal-derived food can be a source of infestation for humans, leading to clinical or sub-clinical disease. Important examples of this pathway include *Toxoplasma* spp. or *Taenia solium*. Foodborne exposure is also possible after environmental contamination, e.g. vegetables or fruit (example *Echinococcus* spp., *Ascaris* spp.). Water can be considered a "food" and as such is another significant exposure pathway for those parasites that are mainly water-borne, e.g. *Giardia* spp. Non-foodborne exposure pathways are common due to the diverse biology and transmission pathways of parasites (including ecto-parasites and protozoa). Infestation of humans can impact on labour force and hereby reduce the productivity of farmers and the amount of food they are able to produce. An important example for this pathway is malaria. Similarly, parasites can infest livestock and – without further spread to consumers – impact on the growth and productivity of food animals, thus reducing the amount of food that would normally have been available from them. Important examples of this exposure pathway are *Theileria parva* but also nematodes and trematodes. In conclusion, parasites have extremely diverse links to food security with direct and indirect impact depending on their biology. While some data on the direct health impact are becoming available (e.g. Torgerson *et al.,* 2013), more work is needed to assess their full impact and role in food security and to set priorities for interventions to assure food security long-term. A good example is the current project GLOWORM (www.gloworm.eu).

11:30 AM - 11:45 AM (15 mins)
Farmer control of gastrointestinal parasites: why they do what they do?
Jacques Cabaret, Chylinski C., Duperray F., Meradi S., Evrard C., Bouilhol M., Berrag B., Sallé G., Nicourt C.
INRA Tours France INRA Ivry France INRA Le Magneraud France Batna University Algeria IVA Hassan 2 Rabat Morocco

Substantial efforts have been made by parasitologists to provide farmers with the technical advice when it comes to the control of gastrointestinal nematodes (GIN) in farm animals. Yet the extent to which the farmers choose to follow the advice of the 'experts' varies substantially. This study explored why farmer adherence to technical advice varies. Data on GIN management was obtained either by questionnaire or semi-directive interviews of sheep, horse and pullet farmers; evaluation of GIN infection was done by standard faecal egg counts. We hypothesized that the human health belief model by Abraham and Sheeran (2007) would provide a useful bases upon which to interpret the GIN management practices of animals. The model is based on: a) threat perception, and b) behavioural evaluation on management of GIN. The results show that threat perception varies between different husbandries of domestic animals: GIN are considered as greater problem in meat sheep production than in horses or free-range pullets. The behavioural evaluation is restricted to the choice of the drug and the moment for treatment. This evaluation is either decided by the farmer, the veterinarian, both, or an integrative contractor. In conclusion, the Abraham and Sheeran model is useful to understand farmers' management of GIN. Farmers have a much wider grid of evaluation for GIN threat and their possible management strategies than the simple technical advice proposed by parasitologists. These results provide insights into considerations that should be taken into account when technical based measures are proposed.

11:45 AM - 12:00 PM (15 mins)

Aquatic Food security: the role of parasites in seafood production

Rachel Norman
University of Stirling, Stirling FK9 4LA

Food security is a global issue as climate change and growing populations put pressure on the planet's resources. There is lots of interesting and important research being carried out to look at issues in food security, however the role of seafood is often ignored. This is despite the fact that Aquaculture is the fasted growing agricultural sector. It now accounts for 50% of the fish eaten globally and is one area in which there is the potential to expand our production without the same limitations of land use that terrestrial systems suffer from. The University of Stirling has recently formed a Centre for Aquatic Food Security which builds on significant expertise in Aquaculture. In this presentation we will look at the role of seafood (both wild capture fisheries and aquaculture) in food security and, in particular, how parasites impact on that.

12:00 PM - 12:15 PM (15 mins)

Forecasting *Nematodirus battus* disease incidence by modelling known hatching dynamics *

Owen Gethings, Eric Rene Morgan, Jan van Dijk, Sian Mitchell
University of Bristol, Woodland Road, Bristol, BS8 1UG

Nematodirus battus is a significant threat to sheep production, with the mass spring hatch of larvae leading to high morbidity and mortality among 6 - 12 week old lambs. Recent climate warming has led to the significant advancement of spring, enabling *N. battus* larvae to begin to hatch earlier in the year. Data on clinical cases of nematodirosis were used to examine trends in hatching date and disease severity within and between years. Disease data pointed to earlier predicted appearance of larvae on pasture in recent years in all regions examined ($p<0.001$). This advancement was associated with a decrease in disease incidence in the South West of England ($p=0.002$), and an increase in Scotland ($p<0.001$). The date of predicted hatch was positively correlated with the number of case submissions in the South West of England ($p=0.007$), suggesting delayed hatching led to an increase in disease incidence. All regions except Scotland revealed positive correlations between the predicted percentage of lambs at risk and the date of predicted hatch ($p<0.001$). Climate warming is predicted to decrease disease risk in some locations by increasing phenological mismatch, whilst increasing disease in other locations. In addition, the use of real time air and soil temperature has enabled the predictive modelling of *N. battus* larvae. Mean March air temperature predicted larval hatch at local ($r=0.661$, $p=0.030$) and regional ($r=0.761$, $p=0.011$) levels. These predictive models have the potential to improve the timing of treatment or the implementation of evasive grazing techniques.

12:15 PM - 12:30 PM (15 mins)

Improved use of abattoir information to aid the management of liver fluke in cattle and sheep *

Stella Mazeri, I. G. Handel, B. M. deC. Bronsvoort, N. D. Sargison
The Roslin Institute, University of Edinburgh Easter Bush Midlothian EH25 9RG Scotland, UK

The incidence and distribution of fasciolosis in the UK has been increasing in the last decade while the timing of acute disease is becoming more variable. Meanwhile control is proving increasingly difficult due to changing weather conditions, increased animal movements and developing anthelmintic resistance. Moreover, the lack of information on the accuracy of meat inspection and available liver fluke diagnostic tests hinders effective monitoring of disease prevalence and treatment. This project explores the value of slaughterhouse data in understanding the changing epidemiology of fasciolosis, identifying sustainable control measures and estimating the effect of infection on production parameters using data collected at one of the biggest cattle and sheep abattoirs in Scotland. Knowledge of the diagnostic sensitivity and specificity of abattoir meat inspection, faecal egg counts, copro-antigen ELISA, serum antibody ELISA and liver necropsy is essential in order to accurately explore the questions stated above. For this reason a Bayesian no gold standard analysis was carried out on results of the aforementioned tests on abattoir samples collected over two sampling periods: during the summer of 2013 and winter of 2014. Preliminary results from this analysis will be presented.

Poster abstracts

The myxoma virus compromises both macro- and micro-parasite immunity in rabbits (P1)

Brian Boag 1, Alex Hernandez2, Isabella Cattadori3, Roy Neilson1
1 The James Hutton Institute, Invergowrie, Dundee, Scotland, DD2 5DA, 2 Rutger University, New Brunswick, New Jersey 08901, USA 3 CIDD, The Pennsylvania State University, University Park, PA 16802, USA

The myxoma virus which has been widespread in wild rabbit populations for c.60 years initially caused high mortality but subsequently, due to the virus becoming less virulent and a build up of resistance in the rabbit, populations had increased significantly until the mid 1990s when a new calici virus (Rabbit Haemorrhagic Disease Virus, RHDV) became widespread. A long term study of rabbits and their parasites and diseases has indicated that myxomatosis can also indirectly increase both helminth and protozoan parasites which could have an additional adverse impact on the health of infected rabbits. The data shows that myxomatosis infected rabbits had mean *Trichostrongylus retortaeformis* worm burdens of 2819 compared with 941 in myxomatosis free animals and that mean faecal nematode egg and coccidia oocyst / g counts were 911 and 73665 respectively in myxomatosis infected animals compared with 427 and 31952 in myxomatosis free animals. The probably reason for the increase in parasites in myxomatosis infected rabbits is that the myxoma virus has the ability to compromises both Th1 and Th2 immunity processes. If the direct impact of the myxoma virus continues to wain then the detrimental indirect impact of the myxoma virus may become relatively more important.

Loop-mediated isothermal amplification (LAMP) reaction in diagnosis of Toxoplasmosis (P2)

Amna Bajwa*1, Saher Islam1, Wasim Shehzad1 Imran Rashid2, Kamran Ashraf2, Haroon Akbar2
1 Institute Of Biochemistry And Biotechnology, University Of Veterinary And Animal Sciences Lahore, Pakistan. 2 Department Of Parasitology, University Of Veterinary And Animal Sciences Lahore, Pakistan.

Toxoplasmosis is a zoonotic and widespread disease, caused by an intracellular, obligate protozoan parasite, *Toxoplasma gondii*. It is a ubiquitous, single species parasitic protozoa affects almost all vertebrates around the world. The cats are definite hosts for *T. gondii*. It can be transmitted to intermediate hosts (e.g., birds and humans) by direct or indirect ingestion of oocysts. The accurate diagnosis is a crucial step using various available methods. Traditional microscopy is used as the first examination, having inherent limitation of diagnosis due to structural similarities with other parasites, serological methods are given an upper hand for further confirmation of the parasite infection. Molecular based approaches like PCR, nested PCR and real-time PCR are also being used but these are expensive and time consuming to use for routine tests. Moreover, sophisticated instrumentation is required for them, makes these impracticable in third world countries. Recently a novel technique of nucleic acid amplification, Loop mediated isothermal amplification (LAMP) has been introduced. The approach is robust, cost effective and reliable that proceeds at isothermal conditions using strand displacement reaction. It provides high amplification and specificity to a target gene. Presence of white precipitates indicate the nucleic acid amplification by LAMP assay. This technique has revolutionized the field of diagnosis by providing early, accurate and quick results. We highlight here the global threat of toxoplasmosis along with the utility of LAMP for diagnosis of the infection. Furthermore different diagnostic approaches are discussed in comparison with LAMP in order to evaluate its utility for diagnostic purpose.

The effect of antihelminthic treatment on schistosome morbidity and immunity (P3)

Naomi Laura Fulton1,2, Paolo Motta3, Norman Nausch1,2, Nicholas Midzi4, Takafira Mduluza5 Francisca Muatpi1,2
1 Institute of Immunology & Infection Research, 2 Centre for Immunity, Infection & Evolution, School of Biological Sciences, University of Edinburgh, Ashworth Laboratories, King's Buildings, West Mains Rd, EH9 3JT, Edinburgh, UK, 3 Roslin Institute, University of Edinburgh, Edinburgh, UK, 4 National Institute of Health Research, P.O. Box CY 573, Causeway, Harare, Zimbabwe, 5 University of Zimbabwe, Biochemistry Department, P.O. Box MP167, Mount Pleasant, Harare, Zimbabwe

Schistosomiasis is a major human parasitic disease affecting over 200 million people in Africa, the Middle East and Latin America. More specifically 400 million people are at risk of infection with *Schistosoma haematobium* which causes urogenital schistosomiasis. Currently the World Health Organisation provides de-worming strategies using the drug

praziquantel (PZQ) for school aged children to prevent the development of severe morbidity. The aim of this study was to measure the effect of antihelminthic treatment with PZQ on morbidity markers in school aged children. Circulating levels of morbidity markers (CHI3L1, CRP, ferritin, resistin and SLPI) were measured in 168 children up to the age of ten years old, living in an area endemic for *Schistosoma haematobium* in Zimbabwe, before and after treatment with PZQ. The current study findings suggest that CRP and CHI3L1 are correlated with infection with schistosomiasis. The involvement of these two biomarkers in the biological processes of initial inflammation suggests that they are predictive of the early stages of schistosome-related pathology. Studies are currently underway to determine how these morbidity markers are affected by treatment.

DRBD5 is a stage-specific mRNA repressor in slender form *Trypanosoma brucei* (P4)

Eva Rico Vidal1, Lucy Glover2, David Horn2, Keith R. Matthews1
1 University of Edinburgh, 2 University of Dundee

During their differentiation from slender parasites to stumpy forms in blood, African trypanosomes down-regulate the expression of many genes as they become quiescent in preparation for transmission. However, a small subset of genes is up-regulated, particularly the ESAG9 family whose mRNAs are highly elevated in intermediate and stumpy forms. ESAG9 proteins are apparently secreted into the bloodstream, although their function is unknown. The regulation of ESAG9 expression has been characterised, demonstrating the importance of the 3'UTR in silencing gene expression in slender forms and allowing expression in stumpy forms. To understand developmental gene regulation between slender and stumpy forms we exploited a genome-wide reverse genetic screen in which a *T. brucei* RNAi library was transfected into parasites expressing a neomycin resistance gene controlled by the ESAG9 3'UTR. Under elevated geneticin selection, the screen selected slender form parasites where ESAG9 3'UTR-mediated gene silencing was reduced, generating increased neomycin expression and therefore increased geneticin resitance. This screen successfully identified the RRM-motif containing protein DRDB5 as a negative regulator operating on the ESAG9 3'UTR in slender forms. This was reproduced by individual DRBD5-RNAi in our neomycin reporter line, and the opposite effect was observed when DRBD5 was over-expressed. However, DRDB5 does not seem to be the only negative regulator acting upon ESAG9 mRNA expression in slender forms since DRDB5 silencing was not sufficient to alleviate the repression of endogenous ESAG9 transcripts. Nonetheless, DRBD5 is strongly developmentally regulated, its absence in stumpy forms likely allowing the expression of some or many stumpy-specific transcripts.

Exploring immune gene expression relative to sheep resistance against *Haemonchus contorus*: A story of sex (P5)*

Jacques Cortet, Caroline Chylinski, Christelle Grisez, Françoise Prevot, Philippe Jacquiet, Jacques Cabaret
INRA Tours France, Ecole Nationale Vétérinaire de Toulouse France

Mounting evidence suggests that a sex effect exists in the capacity of hosts to resist parasitic infections, whereby the males are consistently found to be the 'weaker sex'. Using sheep infected with the gastrointestinal nematode *Haemonchus contortus* as a model, this study explored whether the sex effect could be attributed to differential expression of genes associated with the protective response. An experiment using 49 *H. contortus* infected adult Martinik Blackbelly sheep (24 ewes, 25 rams) found low levels of infection in the males (mean 1664 EPG), but near complete resistance in the females (mean 2 EPG). Eight of the most extreme sheep were selected for further analyses using quantitative RT-PCR i.e. the most resistant and susceptible males and females. Ten immune genes previously implicated in the resistance of Blackbelly lambs were compared in the abomasal lymph nodes and mucosa tissues. Data analyses focused on qualitative trends and networks. The results show the males had a stronger expression of the Th2 cytokines (IL-4, IL-5, IL-13) typically associated with resistance. The females had a greater expression of genes associated with the mucosal defenses, including the secretion of lectins (Intelectin 2, Galectin 15) and epithelial repair (Trefoil factor 3). Furthermore, up-regulation occurred most frequently in the abomasal lymph nodes for the males but the abomasal mucosa in females. This is the first known attempt to explore the sex effect in adult sheep resistance against gastrointestinal nematodes. The results provide interesting insights and potential new avenues for future research into this phenomenon.

Description of *Rhadinorhynchus dorsoventrospinosus* (Acanthocephala: *Rhadinor-hynchidae*) from the red spot emperor *Lethrinus lentjan* with new host and locality records in Saudi Arabia (P6)

Ali AlGhamdi,
Department of Biology, College of Science, Al Baha University, Al Baha, Saudi Arabia.

Adult worms of *Rhadinorhynchus dorsoventrospinosus* (Acanthocephala: *Rhadinor-hynchidae*) were collected from the small intestine of the red spot emperor *Lethrinus lentjan* (family Lethrinidae) from locations along the Red Sea at Jeddah City, Saudi Arabia. Twenty three out of 70 fish specimens (32.9%) were found to be naturally infected. The parasite was described using Zeiss microscopy and a scanning electron microscope (SEM). Light microscopic studies revealed that the adult worm possessed a proboscis which was long, cylindrical with a uniform width measured 0.44 +/- 0.02 (0.38-0.46) mm in length and 0.1 +/- 10.02 (0.09-0.15) mm in width. Proboscis hooks observed by scanning electron microscopy were large, uniform in size (14-16 rows of 26 hooks each) with a row of longer hooks at the base. Comparison between the present described species and four species of the same genus was done, it was observed that there was only one comparable species, *R. dorsoventrospinosus* resembled the present parasite in the general morphology and differed from others, so the present studied species is classified as *R. dorsoventrospinosus* with new host and locality records.

SUMOylation of chromatin-associated proteins by the E3 Ligase SIZ1 positively regulates VSG transcription in trypanosomes (P7)

Miguel Navarro, Diana López-Farfán, Jean-Mathieu Bart, Domingo I. Rojas-Barros
Instituto de Parasitología y Biomedicina "López-Neyra" CSIC, Consejo Superior de Investigaciones Científicas (Spanish National Research Council) Avda. del Conocimiento s/n 18016 Granada Spain

Bloodstream African trypanosomes avoid the host immune response by switching the expression of their surface proteins between Variant Surface Glycoprotein (VSG), only one of which is expressed at any given time. Monoallelic transcription of the telomeric VSG Expression Site (ES) locus localizes to a unique nuclear body named the ESB. Most published work focus in silencing mechanisms of inactive VSG-ESs, however the mechanism involved in transcriptional activation of a single VSG-ES remain totally unknown. Here, we identify a highly SUMOylated focus (HSF) in the nucleus of the bloodstream form that co-localizes with the ESB and the active VSG-ES locus. SUMOylation of chromatin-associated proteins was detected at the active VSG-ES promoter region, but not in silent promoters, suggesting that it is a distinct feature of VSG-ES monoallelic transcription. We identified the SIZ1/PIAS1 SUMO E3 ligase required for SUMOylation detected in the active VSG-ES chromatin. SUMOylation of the VSG-ES chromatin-associated proteins by TbSIZ1 was found to be essential for efficient recruitment of RNA polymerase I to the VSG-ES promoter and for active transcription. Likewise, depletion SUMO-conjugated proteins by either TbUBC9 or TbSUMO knockdown showed a reduction of VSG-ES transcription and pol I occupancy, suggesting that SUMOylation is important for active VSG-ES transcription. Furthermore, RNA pol I largest sub-subunit TbRPA1 is SUMOylated in a TbSIZ-depending manner, confirming a crucial function for SUMO in VSG-ES expression. These data show a positive mechanism associated with VSG-ES monoalleic transcription via post-translational modification.

Tissue models for studying host-parasite interactions with salmon lice *Lepeophtheirus salmonis* (Copepoda, Caligidae) (P8)*

Hazel McDonald1, Andrew P Shinn1, Kim D Thompson1, K Fiona Muir 1, Sean J Monaghan1, Carol M McNair1, Randolph H Richards1, David P Knox2, Scott Hamilton2, David Asper3, James E Bron1
1 Institute of Aquaculture, University of Stirling, Stirling, FK9 4LA, 2 Moredun Research Institute, Pentlands Science Park, UK, EH26 0PZ, 3 Zoetis Aquaculture Biologics Research, Saanichton, BC, V8M 1Z8, Canada

The sea louse, *Lepeophtheirus salmonis,* causes serious problems in salmon aquaculture. Use of integrated pest management strategies using veterinary medicines and a range of farm management tools has provided some control; but a commercially viable vaccine to reduce infection would be highly advantageous. Current methodologies for study of vaccines and host immune responses involve the use of large numbers of fish; however, the development of *in vitro* Atlantic salmon tissue models would assist research on host-parasite interactions and contribute to reduction of animals in research. Fish scale models could provide the structural support and tissue stability needed for maintenance of sea lice but there are limited previous studies into their use. Investigations into the culture of scale-associated epithelial cells were undertaken, which examined various parameters in order to optimise culture conditions. Scales were taken from seawater Atlantic salmon, plated in plastic tissue culture dishes under variable conditions and incubated with medium under CO_2. From initial experiments, it was observed that scales from the caudal region of fish provided successful outgrowth of epithelial cells when placed in groups under standard tissue culture conditions and the use of certain extracellular matrix proteins were also shown to enhance outgrowth when used as a coverslip coating. Development of a viable culture technique could allow limited maintenance of sea lice larvae *in vitro* and provide a platform for investigation of localised host-parasite interactions, however, the conditions required to maintain sea louse viability

must also be considered in developing a final model.

***In vitro* inhibitory effect of Iberian species of the *Cystoseira* genus upon *Leishmania infantum* (P9)**

Carolina Bruno de Sousa1,2, Macridaquez J1, Brito L1, Oliveira M1,2, Florindo C3, Alberício F4, Campino L2, Barreira L1, Custódio L1, Varela J1

1 Centre of Marine Sciences, University of Algarve (UAlg), Faro, Portugal, 2 Unidade de Parasitologia Médica, Instituto de Higiene e Medicina Tropical (IHMT), Universidade Nova de Lisboa (UNL), Lisboa, Portugal, 3 Departamento de Ciências Biomedicas e Medicina, UAlg; Regenerative Medicine Program, Departamento de Ciências Biomédicas e Medicina, UAlg, Faro, Portugal; Centre for Molecular and Structural Biomedicine, CBME/ IBB, UAlg, Faro, Portugal, 4 Institute for Research in Biomedicine, Barcelona Science Park, Barcelona, Spain; CIBER-BBN, Networking Centre on Bioengineering, Biomaterials and Nanomedicine, Barcelona, Science Park, Barcelona, Spain; School of Chemistry, University of KwaZulu-Natal, Durban, South Africa; University of Barcelona, Department of Organic Chemistry, Barcelona, Spain.

The search for new, more effective and less toxic drugs to treat leishmaniasis is undoubtedly relevant. The search for natural products with pharmacological properties has increased in the last few years, particularly the discovery of molecules isolated from marine organisms, holders of a considerable diversity of secondary metabolites with unique structures and functions. Recent studies have shown that several seaweed species may contain biochemicals with anti-leishmanial activity. This study evaluates the *in vitro* activity of hexane, dichloromethane and methanol extracts of *Cystoseira* brown algae (*C. baccata, C. barbata, C. compressa, C. humilis, C. nodicaulis, C. tamariscifolia* and *C. usneoides*) against *Leishmania infantum* promastigotes and mammalian macrophage cells using the MTT assay. To unravel the mode of action of bioactive extracts, parasite morphology, DNA fragmentation, phosphatidylserine externalization, reactive oxygen species production and mitochondrial membrane potential alterations were evaluated. Around 60% of the extracts tested were active against promastigotes. The best results were obtained with the hexane (20.5±1.9µg/mL) and dichloromethane (21.1±3.1µg/mL) extracts of C. baccata and the hexane (11.8±0.3µg/mL) extract of *C. tamariscifolia*, which combine low IC50 values with selective cytotoxicity on the parasite as compared with its effect on the THP-1 cell line. These results suggest that Iberian *Cystoseira* algae contain compounds with anti-leishmanial activity. Further research on the intracellular activity of bioactive extracts as well as bioassay-guided fractionation are in progress.

The efficacy of plant-derived cysteine proteinases as anthelmintics for intestinal nematode infections in small and large mammalian hosts (P10)

Jerzy Behnke, Levecke B., Buttle D., Duce I., Vercruyssse J

Department of Virology, Parasitology and Immunology, Ghent University, Faculty of Veterinary Medicine, Merelbeke, Belgium, Department of Infection & Immunity, University of Sheffield Medical School, Sheffield, S10 2RX, UK. School of Life Sciences, University of Nottingham, University Park, Nottingham, NG7 2RD, UK.

Plant derived cysteine proteinases attack the cuticle of parasitic nematodes, digesting it from the exterior, weakening its structure and eventually causing the target nematodes to die as the cuticle bursts through the force of the high internal hydrostatic pressure. *In vivo*, treatment with a supernatant from papaya latex has been shown to expel more than 90% of intestinal lumen resident *Heligmosomoides bakeri* and *Trichuris muris* in mice, with comparable efficacy against *Haemonchus contortus* in sheep. In this paper we will briefly review the earlier work on mouse models and in sheep, and present novel data showing that single-dose treatment of pigs with the same supernatant extract of papaya latex as that used for the mouse and sheep trials, has higher efficacy against *Trichuris suis* than a standard dose of the synthetic anthelmintic albendazole.

Isolation and Identification of Culicoides Species and *Culicoides Imicola* from Epizootic Hemorrhagic Disease Seropositive Areas in Northern Jordan (P11)

Rami Mukbel, Abd Almajeed, M. Alajlouni, Ahmad Al-Majali.

Department of Basic Veterinary Sciences, Faculty of Veterinary Medicine, Jordan University of Science and Technology, Irbid, Jordan.

Culicoides are small blood sucking biting midges attacking mammals (including human) and birds. Culicoides act as a vector for transmitting a large number of viruses, protozoa and filarial worms to livestock. Most important cattle viral diseases in Jordan transmitted by these midges are the Epizootic Hemorrhagic Disease (EHD), Blue Tongue (BT) and Bovine Ephemeral Fever (BEF). The aim of the present study is to determine the presence of Culicoides species and *C.*

imicola in Northern Jordan. The Culicoides were trapped using CDC light traps, morphologically identified and then confirmed using PCR to amplify ITS1 Nuclear ribosomal DNA (rDNA) gene. Our results documented for the first time the presence of *C. imicola* in at least three areas in the North of Jordan (Middle Jordan valley (DeirAlla), Northern Jordan valley, and Bani-Kenanah). Relative risk maps were made using GIS depending on the association between the *C. imicola* peak activity and climatic variables. The collected insects were tested for the presence of EHD, BTV and BEFV viruses using qRT-PCR. None of the samples were positive for the tested viruses.

ELISA development for the identification of *Neospora caninum* persistently infected carrier cattle: employing a recombinant truncated NcSRS44 protein (P12)*

Stefano Guido1, Jackie Thomson1, Elspeth Milne2, Elisabeth Innes1, Frank Katzer1
1 Moredun Research Institute, EH26 0PZ, Scotland, 2 The University of Edinburgh, EH25 9RG, Scotland

Neospora caninum emerged as a major cause of bovine abortion and neonatal mortality. Despite numerous available ELISA tests, the identification of persistently infected animals remains unreliable due to stage specific antigen expression by the parasite. SRS (SAG-1 Related Sequences) represents a family of genes encoding surface antigens expressed in a stage-specific manner by both *N. caninum* and *Toxoplasma gondii*. Since the genomes of these two apicomplexans share many protein-coding genes, *T. gondii* was used as a model for the identification of putative antigen genes that are expressed during the bradyzoite stage. The *N. caninum* ortholog (NCLIV_040495) of *T. gondii* SRS44 (TGME49_064660) that encodes CST1 was obtained by mining the assembled and annotated genome of *T. gondii*. CST1 is a stage specifically expressed glycoprotein that is an essential component of *T. gondii* tissue cyst wall of the parasite. Expression of NcSRS44 truncated recombinant proteins was successfully achieved in bacterial systems. The immunoreactivity of these proteins was assessed by Western-Blot. The NcSRS44 proteins reacted with known positive sera and they were also recognised by sera from *N. caninum* infected animals that tested negative in commercial ELISAs. The purified truncated recombinant NcSRS44 proteins will be evaluated in an ELISA format to assess their specificity, sensitivity and accuracy in identifying persistently infected cattle.

A forward genetic approach identifies genes involved in *P. falciparum* erythrocyte invasion (P13)

Susana Campino1, Michel Theron1, Magnus Manske1, Kelda Gould, Eleanor Drury1, Daniel, Alcock 1, Bronwyn MacInnis1, Taane G. Clark 2, Dominic P. Kwiatkowski1, Julian C. Rayner1
1 Wellcome Trust Sanger Institute (WTSI), Hinxton, Cambridge, 2 London School of Hygiene and Tropical Medicine, London

The malaria parasite *Plasmodium falciparum* (Pf) can use a complex repertoire of ligand- receptor combinations to invade host erythrocytes. Field and laboratory parasite strains vary in their preferential route of invasion and in their ability to switch pathways. Whilst some of the pathways are known, several are yet to be explored. Genetic studies using Pf clones with different erythrocyte invasion profiles and genetic backgrounds can assist with identifying new pathways. We investigated the invasion profile of a progeny from a cross between 7G8 (Brazil) and GB4 (Ghana) clones and pursued a linkage analysis using whole genome sequence data. We identified two loci associated with erythrocyte invasion, containing genes previously related to invasion as well as new candidates for further exploration.

How do FcμR receptors for IgM influence *Plasmodium falciparum* malaria? (P14)

Katy Lloyd, Blundell, P., Urban, B.C., Pleass, R.J.
Molecular Biochemistry and Parasitology Department, Liverpool School of Tropical Medicine, Liverpool, United Kingdom, L3 5QA

Malaria is a devastating infectious disease caused by *Plasmodium* parasites, where *Plasmodium falciparum* causes ~660,000 deaths annually mainly in children under the age of five. Immunity to malaria is poorly understood, and the parasite's ability to evade and even inhibit immune responses further impedes this understanding. Human IgM contributes to the pathogenesis of severe malaria disease by binding non-specifically to Duffy-binding-like domains (DBLs) expressed by a variety of malarial proteins. How the binding of human IgM to DBLs contributes to immune evasion and pathogenesis is largely unknown, although recent research has shown that IgM binding camouflages parasites from the neutralizing effects of parasite-specific IgG. Understanding why parasites express IgM-binding proteins will provide an insight into malarial immune evasion mechanisms and may help in the development of more effective malaria vaccines. One possibility is that DBLs may bind directly to human IgM opsonised on the surface of

human lymphocytes via Fcμ-receptors (FcμR) that are expressed by these cells. Here we show, by flow cytometry, that DBLs can bind directly to human lymphocytes from healthy donors, and to cell lines expressing FcμRs. We found no direct effect of binding on lymphocyte proliferation, inhibition of proliferation, or apoptosis. To further investigate how the binding of DBLs to lymphocytes may affect the biological and immunological functions of lymphocytes, we sought to unravel the function of the FcμR on these cells. Our initial data suggested a role for FcμR in IgM homeostasis, and we are currently investigating whether the binding of DBLs affect this mechanism.

Ultrastructure and molecular characteristics of a new species of Glugea (Microsporidia: *Glugeidae*) infecting the intestinal wall of *Cephalopholis hemistiktos* from the Red Sea in Saudi Arabia (P15)

Abdel-Azeem Abdel-Baki12, Saleh Al-Quraishy1, Carlos Azevedo1,3
1 Zoology Department, College of Sciences, King Saud University, Riyadh, Saudi, 2 Zoology Department, Faculty of Science, Beni-Suef University, Egypt, 3 Laboratory of Cell Biology, Institute of Biomedical Sciences Abel Salazar (ICBAS/UP), Porto, Portugal

A new microsporidian that infects the marine teleost fish, *Cephalopholis hemistiktos* Rüppell, 1830 that are caught in the Red Sea in Saudi Arabia is described here. This parasite invades the intestinal wall forming white, cyst-like structures containing numerous spores. The prevalence of the infection was 10% (10/100). Mature spores ovoid to pyriform, about 5 µm long and 2.2 µm wide. Polar filament is isofilar and forming 26-29 coils in three rows at the spore's posterior pole. Molecular analysis of the rRNA genes, including the ITS region, and phylogenetic analyses using maximum likelihood were performed. The ultrastructural characteristics and phylogenetic analyses support the recognition of a new species.

The Use of Non-laboratory Animals in Gut Microbiome Studies (P16)*

Emily Pascoe1,2, Géraldine Bastien1, Heidi C Hauffe1, Sarah E Perkins1,2
1 Fondazione Edmund Mach di San Michele all'Adige, Via E. Mach, 1, 38010, San Michele all'Adige (TN), Italy, 2 Cardiff School of Biosciences, The Sir Martin Evans Building, Museum Ave, Cardiff, CF10 3AX

The gut microbiota plays an important role in host health and host-parasite interactions. The composition of the enteric microbiome is influenced by host characteristics; for instance age and genetics, as well as environmental factors such as diet. Much of the information gained on the gut microbiome has been achieved from human studies and laboratory models. However there is still much that can be learnt regarding the enteric microbiota, in particular the interactions that may occur with the parasitic helminth community of the gut. Non-laboratory animals have much to offer the scientific community in terms of how the natural gut microbiota is structured and influenced by interacting factors. Aside from representing greater genetic variation and a natural diet, wild animals exhibit intuitive behaviours. Furthermore non-laboratory species are more likely to possess natural helminth populations in the gut, which is of particular interest when investigating the effect of parasites on gut microbiota. Here, we review the literature to provide an overview of the current research on the microbiota of non-laboratory animals and the driving force behind the choice of certain animal species for study. We describe the variation found between wildlife species in terms of their microbial community and the underlying factors that cause this variation, the role of parasitism in shaping the microbiota and the insight that can be gained from a community ecology style analysis.

Gastrointestinal parasites of exortic Sahelian goats in Sokoto, Nigeria (P17)

Ibrahim Anka Abubakar, Halliru Amadu, Kassu Mansur, Abdullahi Sanusi, Anka Buhari, Kabiru Tambuwal
Department of Parasitology and Entomology, Faculty of Veterinary Medicine, Usmanu Danfodiyo University, PMB 2346 Sokoto, Nigeria.

Little information is available on the gastrointestinal parasites of exotic trade Sahelian goats in Nigeria. Exploratory survey was undertaken to determine the types and pattern of gastrointestinal parasites of trade Sahelian goats in relation to country of origin, age (young: < 1 year; adult: 1 to 4 years and old: > 4 years), sex and season (dry and wet). The investigation was carried out at the goat's resting station located at the vicinity of Usmanu Danfodiyo University, Sokoto, between March and December, 2013. Exploratory data analysis was used to analyse the data based on percentages (%). Faecal samples were collected per rectum separately from each animal and subjected to simple flotation technique for oocysts and nematode eggs detection. Findings showed that all the 362 Sahelian goats originated from Republic of Niger. The number of parasites encountered overall, with their proportions were, *Eimeria* species (155; 42.8%), gastrointestinal strongyles (275; 76%) and *Trichuris* species (1; 0.3%). The highest infection rates recorded age

wise were 53.7% (old goats) as single and 36.9% (adult goats) as dual infections; and 44.1% and 79.3% (old goats) for *Eimeria* and stronglyle parasites, respectively. Sex wise, infection rates were generally similar irrespective of overall infection status (single and dual) or specific type of infection (*Eimeria* and strongyle). Seasonal data however markedly skewed towards the wet season, both in relation to overall infection status and specific type of infection. Our findings show that gastrointestinal parasites infections are common in trade Sahelian goats imported to Nigeria.

Identification of the α-subunit of AMP-activated protein kinase and characterization of the entire complex in *Trypanosoma brucei* (P18)

Chiara Ambuehl, Patricia Graven, Leonardo Scapozza, Remo Perozzo
Pharmaceutical Biochemistry Group, School of Pharmaceutical Science,University of Geneva and Lausanne,Quai Ernest-Ansermet 30, 1211 Geneva 4, Switzerland

AMP-activated protein kinase (AMPK) is a heterotrimeric complex well conserved in all eukaryotic which acts as regulatory sensor of the energy status of the cells. Its role is based mostly on sensing the AMP/ATP ratio of the cells and to restore the adenine nucleotide homeostasis [1]. In the blood stream form of *Trypanosoma brucei*, the parasite responsible for the Human African Trypanosomiasis or sleeping sickness, the AMPK complex has neither been completely identified nor characterized to date. Previous investigations on the procyclic form of *T. brucei* allowed the identification of the β and γ-subunits of TbAMPK [2]. Two possible α subunit candidates have been identified by sequence comparison, but further tests using RNAi methods failed to confirm the role of one of the two candidates in procyclic form of *T. brucei* [2]. It is the aim of this project to confirm the β and γ-subunits and to identify the unknown α subunit in the blood stream form of *T. brucei*. As numerous trials using standard immunoprecipitation protocols could not provide any candidate for the α-subunit, we now apply the recently developed proximity-dependent biotin identification (BioID) assay which consist of tagging the β subunit with a modified 35-kDa bacterial biotin ligase (BirA*) harboring a myc-tag [3,4]. This technique will allow the identification of all elements building up the AMPK complex either by affinity purification via the myc-tag, or by immunoprecipitation of all biotin-labelled proteins after incubation of cells with biotin. In parallel, the creation of two polycistronic expression systems containing the β, γ and the 2 putative α-subunit, respectively, will allow us to identify the good candidate. Previous work demonstrated the capacity of this approach to isolate active recombinant AMPK complexes [5]. The characterization of the recombinant TbAMPK will be carried out with respect to its activation, activity and regulation by adenine nucleotides. [1] David Carling, TIBS. 2004, 29(1), 18. [2] Clemmens, C.S. et al., Exp Parasitol. 2009, 123, 250. [2] Roux K.J. et al., J. Cell Biol. 2012, 196, 801 [4] Morriswood B. et al., Eukariot cell, 2013, 12, 356 [5] Neumann D. et al., Protein Expr Purif 2003, 30, 230

Investigating the zoonotic potential of *Ascaris* and *Trichuris* in Ecuador (P19)*

Alexandra Sparks1, H. Meekums3, G. Oviedo4, C. Sandoval4, M.B.F Hawash5, P. Nejsum5, M. Betson3, P.J. Cooper4,6,7, J.R. Stothard,1
1 Parasitology Department, Liverpool School of Tropical Medicine, Liverpool, UK, 2 Institute of Immunology & Infection Research, School of Biological Sciences, University of Edinburgh, Edinburgh, UK, 3 Department of Production and Population Health, Royal Veterinary College, Hatfield, Herts, UK, 4 Laboratorio de Investigaciones FEPIS, Quinindé, Esmeraldas Province, Ecuador, 5 Department of Veterinary Disease Biology, Faculty of Health and Medical Sciences, University of Copenhagen, Copenhagen, Denmark, 6 Centro de Investigación en Enfermedades Infecciosas, Pontificia Universidad Católica del Ecuador, Quito, Ecuador, 7 Centre for Infection, St George's University of London, London, UK

The soil transmitted helminths *Ascaris lumbricoides* and *Trichuris trichiura* infect over one billion people worldwide. They are closely related to A. suum and T. suis respectively, which infect pigs across the globe with high prevalence in extensive and organic farming systems. The extent of natural cross-transmission of *Ascaris* and *Trichuris* species between human and pig hosts is poorly understood. The aim of this study was to determine whether there was zoonotic transmission of *Ascaris* and *Trichuris* in rural Ecuador. In a survey conducted in Esmeraldas Province, *Ascaris* and *Trichuris* prevalence levels in humans (N=132) were 45.5% and 46.2%, respectively, and 34.8% and 10.9%, respectively, in pigs (N=46). Adult *Ascaris* and *Trichuris* were collected from egg-patent humans and pigs by chemoexpulsion. Additional pig worms were obtained in an abattoir. 116 *Ascaris* from humans and 5 *Ascaris* from pigs were genotyped using an ITS PCR-RFLP assay. Genotyping of 81 human *Trichuris* and 74 pig *Trichuris* was conducted using ITS2 and 18S PCR-RFLP assays. Genotyping of adult worms did not provide evidence of cross-transmission of *Ascaris* and *Trichuris* between pigs and humans in this setting. This result is surprising given that most families in the study owned, or lived in close proximity to pigs and contrasts with data from China and Uganda where zoonotic transmission has been found. This may reflect differences in farming practices in different areas. However, as we have only analysed adult worms, *A. suum* and *T. suis*

may still cause zoonotic infection but only reach immature stages in humans.

Schistosoma haematobium and malaria parasite co-infection and effects on haematological parameters (P20)
Olajumoke Morenikeji,Atanda Omotayo, Ituna Eleng, Salawu Tunde
Ogun State, Nigeria

There have been inconclusive debates on the severity of *Plasmodium falciparum* infection in patients co-infected with *Schistosoma haematobium* and vice versa. This study aims at assessing the association between single infection and co-infection status of the two parasites with haematological profiles. A cross-sectional epidemiological survey was carried out on a total of 202 school children between ages 6-18 years (mean age 11.5±2.6 years). Urine and blood samples were collected by standard methods for concurrent microscopic determination of *S. haematobium* and *P. falciparum* respectively. The following haematological parameters; haematocrit, haemoglobin, neutrophils, leucocytes, lymphocytes and eosinophils were also determined. The prevalence of single infection was 52.0 and 59.9 % for *S. haematobium* and *P. falciparum* respectively, while 28.2% individuals were concurrently infected with the two parasites. All haematological parameters except haematocrit (in all single and co-infection) and lymphocytes (in *S. haematobium* infected children) showed no significant differences in the occurrence of normal and abnormal gradients in both single and co-infection status (P>0.05). It can be concluded that an individual with malaria infection may show an improved lymphocyte profile if co-infected with *Schistosoma haematobium*.

Structural and molecular characterization of _Kudoa quraishii_ n. sp. from the trunk muscle of the Indian Mackerel _Rastrelliger kanagurta_ (_Perciforme, Scombridae_) in Saudi Arabia Coasts (P21)
Lamjed Mansour1,2, Abdul Halim Harrath1, Abdel-Azeem S Abdel-Bali1, Suliman Y. AlOmar1
1 Department of Zoology, College of Science, King Saud University PO. Box: 2455, Riyadh, 11451, Saudi Arabia, 2 Unité de Recherche de Biologie Écologie et Parasitologie des Organismes Aquatiques, Département de Biologie, Faculté des Sciences de Tunis, Campus Universitaire, 2092 Tunis El-Manar, Tunis

A new myxozoa; *Kudoa quraishii* n. sp. is reported in the striated muscle of the Indian mackerel *Rastrelliger kanagurta* from the Red Sea and the Arabian Gulf in Saudi Arabia. Mean prevalence of infection is about 20% and varies between localities. The parasite develops whitish and oval or rounded pseudocysts of 0.2-3 mm in the striated muscles of the body. Pseudocysts are filled with mature spores. Myxospores are quadrate in shape in apical view with rounded edges and ovoid in side view. Each spore is formed by four equal shell valves and four symmetrical polar capsules. Polar capsules are pyriforme in apical view and drop-like in side view. Myxospores measurements in µm are 6.14 (5.9-6.34) in width, 5.48 (5.3-5.71) in thickness, and 4.27 (4.1-4.42) in length. Polar capsules measurements in apical view in µm are 2.08 (1.88-2.28) and 1.31 (1.10-1.52) length by width. Molecular analysis based on SSU rDNA gene shows closest association with *K. amaiensis* and *K. kenti* with respectively 98% and 97.2% of similarities.

Strongyloides strikes back: Strongyloidiasis as a cause of duodenal obstruction in the United Kingdom (P22)
Kartik Kumar, Olga Sotulenko, Geneivieve Hazarika, Manoj Nair, Lee Dvorkin
North Middlesex University Hospital NHS Trust, Sterling Way, London, N18 1QX, UK

Background: Duodenal obstruction is an infrequently documented complication of Strongyloidiasis. We present a case of duodenal obstruction secondary to Strongyloides and consider this in the context of the published literature. Case: A 76 year old Afro-Caribbean man presented with a two week history of vomiting, worsening constipation, abdominal distension and recent weight loss. Examination revealed epigastric tenderness and a succussion splash. Abdominal CT showed marked gastric and duodenal dilatation but no obvious cause for obstruction. Following decompression with a nasogastric tube, duodenal biopsies revealed abundant worms suggestive of Strongyloidiasis. Stool microscopy confirmed infection with *Strongyloides stercoralis* (rhabditiform). A subsequent screening test for HTLV-1 was strongly positive. It was established that the patient had recently travelled to Latin America. Symptoms improved after treatment with mebendazole. Discussion: Excluding our case, eleven cases of duodenal obstruction secondary to Strongyloidiasis have been documented in the literature between 1979 and 2014. Only one of these eleven cases was reported in the UK: coincidentally it was reported from our own institution. Thus our institution has now seen two patients with Strongyloidiasis-induced duodenal obstruction in the last five years. The clinical picture in both patients was very similar: they presented with symptoms of intestinal obstruction; diagnosis was made following positive duodenal biopsy and stool findings; HTLV-1 co-infection was subsequently confirmed; symptoms improved with anthelmintics. Therefore this

highlights the importance of considering Strongyloidiasis as an unusual but nevertheless important potential cause of duodenal obstruction in the UK, especially in the context of confirmed HTLV-1 infection.

Organisation of cathepsin cysteine proteases within the *Fasciola hepatica* genome (P23)

Krystyna Cwiklinski1*, Steve Paterson2, Philippe Dufresne3, John Dalton4, Diana J. Williams1, Jane E. Hodgkinson1
1 Department of Infection Biology, IGH, University of Liverpool, 2 Centre for Genomic Research, University of Liverpool, 3 Institute of Parasitology, McGill University, Ste Anne de Bellevue, 4 Queen's University, Belfast

The liver fluke, *Fasciola hepatica* is an economically important pathogen of sheep and cattle. It has been described as a re-emerging zoonosis and is a major focus of anthelmintic resistance and vaccine development research. Excretory-secretory proteins, particularly the proteolytic cathepsin cysteine proteases, act at the host-parasite interface and aid survival of the migratory stages, the newly excysted juveniles (NEJ) and adults. These important proteases can be subdivided into several clades/subtypes of cathepsin L and B proteases. To increase our understanding of these proteases and how they can be manipulated for vaccine and diagnostic tools, it is important to analyse the genomic organisation and differential transcription of these proteases. Recently we have generated a 1.5 Gb *F. hepatica* draft genome from a clonal isolate by Illumina sequencing. Following assembly into scaffolds, annotation is underway, generating gene models based on *F. hepatica* transcriptome and Schistosome protein sequence. This study reports mining of the F. hepatica genome to determine the genomic organisation of the cathepsin L and B genes. Comparative transcriptomics was performed for metacercariae, NEJ, juvenile and adult fluke lifecycle stages to determine the relative transcription of the cathepsin and legumain proteases. Cathepsin L proteases are transcribed by the juvenile and adult flukes at significantly higher levels than the other lifecycle stages, whereas the cathepsin B proteases are primarily transcribed by the NEJ. This study highlights the potential of the cathepsin proteases for the continued development of early diagnostic tools and vaccines.

Excreted/secreted *Schistosoma mansoni* microRNAs are found within and outside of extracellular vesicles. (P24)*

Fanny C. Nowacki1, Martin T. Swain1, Alasdair C. Ivens2, Amy H. Buck2 and Karl F. Hoffmann1
1 Institute of Biological, Environmental and Rural Sciences, Aberystwyth University, 2 Centre for Immunity, Infection and Evolution, Ashworth Laboratories, University of Edinburgh

Mature microRNAs (miRNAs) are a class of small non-coding RNA involved in post-transcriptional regulation of target messenger RNAs (mRNAs). Their identification is based on experimental and/or computational approaches and, to date, 30424 mature miRNAs have been characterised in 206 plant, animal and viral species. Studies have found significant numbers of miRNAs in extracellular environments, which paradoxically, also contains miRNA-degrading RNases. It is believed that miRNAs remain protected from RNase activity in these environments by association with proteins or by encapsulation in extracellular vesicles (EV). Therefore, extracellular miRNAs released in one location may be stabilized by RNAse-protective proteins/vesicles to exert transcriptional regulation at distant sites. To identify if *Schistosoma mansoni* releases these gene-expression modulators into their immediate environment, we conducted an RNA-seq investigation of excretory/secretory products (ESP) derived from schistosomula. Fractionation of the schistosomula ESP into EV-enriched or EV-depleted samples was facilitated by ultracentrifugation. Transmission electron microscopic (TEM) analysis revealed exosome-like vesicles. Total RNA extraction was performed and small RNA libraries were created from both samples using a NEBnext multiplex Small RNA Library Set for Illumina NGS platforms. The average number of reads per library was 13,500,000. All reads that mapped to the *S. mansoni* genome (v5.1) were qualified for RNA class and quantified for miRNA identity using bespoke pipelines involving Rfam, mirBase and miRDeep2. Interrogating this dataset revealed 50 known and at most 200 novel miRNA found released by schistosomula. The biological significance of these observations is currently under active investigation in our laboratory.

Genome-wide assessment of *Plasmodium falciparum* infection diversity in West Africa. (P25)

Lee Murray1, VA Mobegi1, SA Assefa1, KM Loua2, DP Kwiatkowski3, A Amambua-Ngwa4, CW Duffy1, DJ Conway1
1 London School of Hygiene & Tropical Medicine, Keppel St, London, WC1E 7HT, 2 National Institute of Public Health, Conakry, Republic of Guinea, 3 The Wellcome Trust Sanger Institute, Hinxton, Cambridge, UK, 4 Medical Research Council Unit, Fajara, Banjul, The Gambia.

Previous studies assessing the within-host diversity and population structure of *Plasmodium falciparum*, have tended to focus on the use of PCR-based genotyping to distinguish between distinct parasite clones. Whilst such assays are able to

characterise the number of alleles present within an infection at certain loci, these approaches are limited by the paucity of loci that are used to identify separate clones and the varying conclusions on the multiplicity of infection seen even within the same samples by different laboratories. Recent work has highlighted that a surprisingly high degree of haplotype relatedness is exhibited by within isolate clones in comparison to haplotypes found within separate *P. falciparum* infections. Within this study, we have used a combination of multilocus microsatellite typing and whole genome sequences from The Gambia and Guinea to assess infection diversity along the north/south transmission gradient of West Africa. Using the sequence data, individual isolate Fws scores were calculated and definitions of clonality were compared with the microsatellite typing. In addition, the Fws metric was computed across the genome as a scan to investigate changes in within-host diversity amongst subsets of predominantly clonal and diverse infections.

Current status of rabbit coccidiosis in Egypt (P26)

Hebat Alla Hussein1, Thabet Sakran1, Gamal El-Shahawi1, Huda El-Fayomi1, Shawky Abo-Alhadid2
1 Zoology Department, Faculty of Science, Beni-Suef University, Egypt, 2 Parasitology Department, Faculty of Veterinary Medicine, Beni-Suef University, Egypt

Rabbit coccidiosis is caused by parasites of the genus *Eimeria*, which are true pathogens that are always present in rabbit farms as they are virtually impossible to eradicate. Coccidiosis causes dramatic economic loss in rabbit farms due to diarrhea, weight loss, destruction and fetal outcome of the disease. A few studies have been published regarding the prevalence of rabbit coccidiosis in Egypt. In the present study, the natural prevalence of coccidian infection among domestic rabbit (*Oryctolagus cuniculus*) in Beni-Suef Governorate, Egypt was investigated. Severe overall prevalence reaching 70% (70/100) was recorded. Eight species of *Eimeria* were detected and identified according to their morphometeric characteristics. Mixed infection with three different species occurred most frequently. *Eimeria intestinalis* and *Eimeria coecicola* were generally the most predominant species. On necropsy, ileum and ceacum are the most influenced portion of the intestine. They are congested oedematus and topped up with watery feces. Histological investigations revealed the endogenous stages in the intestinal mucosa with villous atrophy and enterocyte destruction.

Toxoplasmosis: its scarcity in cats and copro-diagnosis through PCR (P27)*

Amna Arshad Bajwa, Rahim Gul, Muhammad Imran Rashid, Haroon Akbar, Ali Ahamd, Habibun Nabi, Saher Islam, Wasim Shehzad
Lahore, Pakistan

Toxoplasma gondii is an obligate intracellular parasite belonging to the phylum Apicomplexa that infects all warm-blooded animals and birds. The cat as a final host is considered culprits for shedding oocysts in the environment thus contaminating food items of human use and it has been exploited for spreading toxoplasmosis in the media since 1969. In the present study, Two hundred fecal samples from domestic cats were examined for *T. gondii* oocysts by simple microscopy. Only 3 out of 200 samples were found positive for toxoplasmosis, it was more prevalent in adult cats (1.65%) as compared to young ones. Prevalence was also found higher in female cats (2.08%) as compared to male cats. PCR was employed to detect *T. gondii* oocysts from fecal samples. Primers were designed by using Primer-Blast software of NCBI targeting B1 gene of toxoplasma. The reaction was optimized to amplify 529 bp sequence for copro-PCR.

Screening of a plant-derived natural product library for anthelmintic activities. (P28)*

Jennifer A. Edwards1, Martha Truscott1, Robert Nash2, Karl F. Hoffmann1
1 Institute of Biological, Environmental and Rural Sciences, Aberystwyth University, SY23 3UG, 2 PhytoQuest, Plas Gogerddan, Aberystywth, SY23 3EB.

Schistosomiasis is a debilitating parasitic disease, caused by blood flukes of the genus *Schistosoma*. Over reliance of mass drug administration (MDA) programmes on a singular chemotherapy, praziquantel (PZQ), combined with reports of low PZQ efficacy and unsuccessful PZQ treatments put future schistosomiasis control methods into question. In response to this, a necessary step for the long-term control of the disease is the development of new anthelmintics. Based on the anti-parasitic successes of artemisinin related compounds, plant-derived natural products offer a potential rich resource for the discovery of new anthelmintics. To this end, *in vitro* anti-schistosomal (*Schistosoma mansoni*) screening of 47 structurally-related plant-derived compounds was performed. Further to this, the activity of a subset of these 47 compounds towards the newly excysted juvenile (NEJ) life stage of the parasitic liver fluke *Fasciola hepatica* was also assessed *in vitro*. From these assays, six compounds were selected as dual anthelmintic hits with two, compounds

700336 and 700992, selected for further investigations. Both compounds possessed moderate to severe activity towards the schistosomula and NEJ lifecycle stages. After further analysis, compound 700336's activity against the adult life stage of *S. mansoni* was also investigated. Compound 700336-mediated phenotypic and biological affects included reductions in egg output and breaches in tegumental integrity. Confocal laser scanning microscopy also showed that compound 700336 affected the normal formation of eggs within adult females. Findings from this work provide valuable information about the properties of these structurally related plant-derived compounds and demonstrate their potential as novel anthelmintic leads.

Sequencing and analysis of the parasitic nematode *Strongyloides ratti* and its close relatives (P29)

Avril Coghlan, Nancy Holroyd, Bernardo Foth, Jason Tsai, Taisei Kikuchi, Alan Tracey, Sarah Nichol, Karen Brooks, Helen Beasley, Eleanor Stanley, Alejandro Sanchez-Flores, James Cotton, Daria Gordon, Anna Protasio, Vicky Hunt, Nadine Randle, Arpita Kulkarni, Jonathan Wastling, James Lok, Jonathan Stoltzfus, Stephen Doyle, Warwick Grant, Adrian Streit, Mark Viney, Matthew Berriman

Wellcome Trust Sanger Institute, Cambridge, UK, Biodiversity center, Academia Sinica, Taiwan, Forestry and Forest Products Research Institute, Ibaraki, Japan National Autonomous University of Mexico, University of Bristol, University of Liverpool, Max Planck Institute for Developmental Biology, Tübingen, Germany University of Pennsylvania, Philadelphia, USA, La Trobe University, Melbourne, Australia

The genus *Strongyloides* contains about 50 species of obligate gastrointestinal parasites of vertebrates. The best-studied species in the genus is *S. ratti*, a parasite of rats, but the genus also includes *S. stercoralis*, a significant human pathogen, with an estimated 30-100 million people infected worldwide. Here we present the sequencing, annotation and analysis of the genomes of *S. ratti, S. stercoralis,* and their close relatives *S. venezuelensis, S. papillosus* and *Parastrongyloides trichosuri*, parasites of rats, ruminants and Australian possums respectively, as well as an outgroup species, *Rhabditophanes* sp. KR3021. We discuss how analyses of their genome sequences has revealed interesting evolutionary processes, such as dramatic genome contraction via loss of non-coding DNA; widespread intrachromosomal rearrangements; and huge expansions of particular gene families, some of which are likely to represent important adaptations for parasitism. To complement the genomic analyses, we have used RNA-seq to identify similarities and differences between species in their expression patterns in infective larvae and parasitic adults, in *S. ratti, S. venezuelensis* and *S. stercoralis*. Lastly, a combination of genetic mapping and quantitative sequencing of males and females has helped us to assign sequences to chromosomal regions and to illuminate the evolution of an intriguing process: a chromatin diminution event that plays a role in sex determination in *S. papillosus*.

Novel histidine phosphatases in trypanosomatid parasites (P30)*

Amber Lynch*1, Robert WB Brown1, Daniel J Rigden2, Paul McKean1, Michael Ginger1

1 Faculty of Health and Medicine, Division of Biomedical and Life Sciences, Lancaster University, Lancaster, LA1 4YQ, UK, 2 Institute of Integrative Biology, University of Liverpool, Crown St, Liverpool, L69 7ZB

Collectively, histidine phosphatases (HPs) form an ancient, ubiquitous enzyme superfamily. The reaction catalysed by family members is dephosphorylation of a substrate, which is critically dependent upon an active-site histidine. The 'classic' or best known HP is the glycolytic enzyme phosphoglycerate mutase (PGAM), but the substrate preferences and physiological function of most HPs are unknown. Kinetoplastids, including members of the parasitic trypanosomatid family, contain a collection of HPs, most of which are sparsely distributed in prokaryotes and other eukaryotes. Given that adaptation to parasitism is classically associated with streamlining, the function of trypanosomatid HPs appears intriguing. Here we will report on the phylogenetic distribution and molecular characterisation of a paralogous pair of histidine phosphatases in the sleeping sickness parasite *Trypanosoma brucei*. Both paralogues are mitochondrial, but curiously one paralogue appears to be catalytically inactive. We will also report on our progress in trying to generate gene deletion mutants in procyclic form trypanosomes. Our current data suggests that both of these genes are essential.

The development of neuropeptides as transgenic nematicides (P31)

Johnathan Dalzell, Neil D. Warnock, Leonie Wilson, Colin C, Fleming, Aaron G. Maule.

Queen's University Belfast

Plant pathogenic nematodes (PPNs) impose a significant economic burden on plant cultivation efforts worldwide. Recent estimates predict losses across all sectors of approximately $125 billion annually. Conventionally, an integrated approach to PPN management has relied heavily on various nematicides. As environmental concerns rise over the systemic effects

of sustained nematicide use, withdrawal has left a significant shortcoming in our ability to manage this problem and highlights the need for novel and robust control methods. It has been discovered that nematodes can assimilate exogenous peptides through retrograde transport along the chemosensory amphid neurons. These peptides accumulate within cells of the central nerve ring and can elicit physiological effects when released to interact with receptors on adjoining cells. We are harnessing bioactive neuropeptides from the neuropeptide-like protein (NLP) and FMRFamide-like peptide (FLP) families of plant parasitic nematodes as novel nematicides. We have identified numerous discrete neuropeptides that impact on diverse behaviours such as chemosensation, motility, infectivity and stylet thrusting of the root knot nematode *Meloidogyne incognita*, and the potato cyst nematode *Globodera pallida* through RNAi-based gene functional studies and exogenous peptide addition. Transgenic approaches to neuropeptide utilisation as novel nematicides have been developed, and provide significant protection of crop plants.

Putative identification by immuno-blotting of the receptor associated membrane protein-2 (ramp-2) in new world *Leishmania* species. (P32)

Alicia Ponte-Sucre, A.Febres, E. Díaz
Laboratory of Molecular Physiology. IME-UCV Central University of Venezuela

Calcitonin Gene Related Peptide (CGRP) interacts with its receptor (CLR) a member of the 7-TM-G protein coupled receptors. Its pharmacological profile varies depending on its co-expression with either of the Receptor Associated Membrane Proteins (RAMP-) -1, -2 or -3. RAMPs alter the cellular traffic and glycosylation of CLR, thus changing the pharmacokinetics of the resulting receptor. In higher eukaryotes, CLR and RAMP-1 form a complex selective to CGRP. On the other hand, CLR association with either RAMP-2 or RAMP-3 results in a receptor with higher affinity for the neuropeptide Adrenomedullin (ADM). By the use of the capillary-two chamber technique we demonstrated that *L. (V.) braziliensis* expresses a negative chemotactic response when exposed to CGRP (10-8, 10-9 M). A similar response was obtained in parasites exposed to ADM (10-5, 10-7 M). These results suggest that *L. braziliensis* expresses a peptide (~Ramp), sensitive to (higher affinity) CGRP and ADM. Evidence of RAMPs being expressed in lower eukaryotes is rare; nevertheless we explored the identity of the involved receptor. By immunoblotting of cell homogenates of both *L. braziliensis* and *L. (L.) amazonensis* we identified a signal suggestive of the expression of a protein carrying an epitope comparable to amino acids 28 – 166 of human RAMP-2. The results suggest that *Leishmania* parasites express a RAMP related peptide that functions as a unique receptor for both peptides.

Optimizing the inhibition of a uniquely composed *Trypanosoma brucei* F_1-ATPase (P33)

Ondrej Gahura, Hanka Vachova, Brian Panicucci, John Walker, Alena Zikova
Institute of Parasitology, Biology Centre ASCR, Ceske Budejovice, Czech Republic Mitochondrial Biology Unit, MRC, Cambridge, UK

The transition of the parasitic *Trypanosoma brucei* between its invertebrate and vertebrate hosts is associated with substantial bioenergetic pathway changes. While substrate and oxidative phosphorylation (OXPHOS) provide the main source of ATP in the procyclic stage (PS), increased glycolysis of abundant glucose in the bloodstream form (BS) compensates for the absence of OXPHOS - requiring the F_1F_o-ATP synthase to function in reverse to maintain the mitochondrial membrane potential at the expense of ATP. A widespread natural protein inhibitor of F_1F_o-ATPase activity (TbIF1) is expressed in PS, while its ectopic expression is lethal in BS. To characterize TbIF1 inhibition, we isolated the F_1-ATPase from PS by two-step chromatography. Besides the conserved eukaryotic components (α3β3γδε), the complex contains an additional trypanosomatid-specific protein, p18. The previously reported α subunit cleavage was confirmed and modeled to a region presumed to form a loop between the crown and NTP-binding domains, a unique feature of F_1-ATPase in Kinetoplastids. Futhermore, several recombinant TbIF1 mutants were characterized to determine their dissociation constants and oligomerization properties. While the C-terminal deletion of TbIF1 prevents homodimerization, it doesn't disrupt the pH sensitivity as it does in bovine IF1. Importantly, TbIF1 can't inhibit bovine F_1-ATPase and vice versa, strengthening the differences between the parasite and mammals. The established purification of a uniquely composed F_1-ATPase is suitable for structure resolution by X-ray crystallography. Given the non-conventional function of F_1-ATPase in BS and the F_1-TbIF1 binding data, we propose that the structure could be exploited to design specific inhibitors for potential use in therapeutics.

Do different African trypanosome species share quorum-sensing signal responses? (P34)*

Eleanor Silvester, Keith R. Matthews

Centre for Immunity, Infection and Evolution, School of Biological Sciences, University of Edinburgh

Animal African Trypanosomiasis is predominantly caused by the protozoan pathogens *Trypanosoma vivax, Trypanosoma congolense*, and *Trypanosoma brucei brucei*. During mammalian bloodstream infection, *T. brucei* demonstrates pleomorphism. Initially proliferative 'slender' forms dominate the infection, but as parasitaemia increases an accumulation of an unidentified stumpy induction factor (SIF) drives differentiation to a 'stumpy' form. This growth-arrested form is pre-adapted for transmission to the tsetse fly vector. We are currently investigating whether mechanisms of growth control in *T. brucei* are conserved in *T. congolense* and *T. vivax*, this potentially impacting on virulence in single and coinfections. Following our recent identification of a cohort of genes involved in driving stumpy formation in *T. brucei*, we have carried out a bioinformatics analysis of the conservation of these regulators in *T. congolense* and *T. vivax*. This demonstrated that many of the genes are shared between these species, highlighting the potential for shared mechanisms of growth control. To investigate the functional conservation of these genes we have initiated a systematic analysis of the ability of the *T. congolense* or *T. vivax* orthologues to restore stumpy formation in *T. brucei* lines where each gene is silenced by RNAi and thereby unable to respond to SIF. Furthermore, using transgenic *T. brucei* lines that report on stumpy formation via CAT expression, we have begun analysis of the potential for signal cross talk between *T. brucei* and *T. congolense* in coinfections by analysing the effect of conditioned medium from cultured *T. congolense* bloodstream forms.

Molecular targets for plant cysteine proteinases on parasitic nematode cuticles (P35)*

Victor Stephen Njom1, Tim Winks1, Ian Duce2, Mark Dickman3, and David J. Buttle1
1 *Infection and Immunity, 2 Biological and Chemical Engineering, University of Sheffield, 3 School of Biology, University of Nottingham.*

Parasitic nematodes cause enormous public health, agricultural and economic problems worldwide, as pathogens of humans, livestock and crops. Their impact is increasing due to resistance to current anthelmintics. We are developing plant cysteine proteinases as alternatives to currently available anthelmintics. The enzymes digest the outer cuticle of the nematode leading to blistering followed by rupture and death. The nematode cuticle is composed of proteins such as collagens and cuticlins but the specific molecular target(s) of the proteinases have not been identified. There are about 170 collagen genes and 30 cuticlins in the *C. elegans* genome. Our study aims to identify the molecular target(s) and thereby define the mechanism of action of these proteinases. Washed cuticles of *Caenorhabditis elegans* and the murine GI nematode *Heligmosomoides bakeri* were digested with papain, the supernatant was run on SDS-PAGE and individual bands were analysed by LCMSMS. Paramyosin, cuticle globin and actin-related proteins were identified in papain-digested cuticle supernatants. Preliminary data suggest that our approach is a practical one. However, structural cuticle proteins attacked by plant cysteine proteinases have yet to be identified.

cAMP Binding Proteins in *T. cruz*i (P36)

Martin M. Edreira, Bárbara Mc Cormack, Adriana V. Jäger, Javier G. De Gaudenzi, Jesica G. Mild, Daniel Musikant, Sergio Pantano, Daniel L. Altschuler
Departamento de Química Biológica, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Argentina, Instituto de Investigaciones Biotecnológicas, IIB-UNSAM-CONICET, Buenos Aires, Argentina, Institut Pasteur, Montevideo, Uruguay, Department of Pharmacology and Chemical Biology, School of Medicine, University of Pittsburgh, Pittsburgh, PA.

Trypanosoma cruzi has a complex life cycle that involves four morphogenetic stages with various second messenger systems capable of regulating its growth and differentiation through signal transduction cascades. Particularly, cAMP-dependent signaling shown to have a crucial role in the proliferation/differentiation and invasion of the host cell. For long, PKA has been considered as the unique cAMP effector. However, in superior eukaryotes a new effector for this second messenger was recently found: EPAC (Exchange Protein directly Activated by cAMP). The absence of sequences for Epac or Epac-like proteins in *T. cruzi* genome proved that the cAMP-EPAC pathway is not present in the parasite. Although, experimental evidences suggests the existence of PKA-independent pathways in *Trypanosoma*. In order to identify new potential cAMP effectors, we carried out in silico search that identified 27 proteins with putative cAMP binding domains. Phylogenetic analysis made from coding sequence alignments showed that these proteins are segregated into two main branches: one containing protein kinases and the other gathering hypothetical proteins with no assigned function. Putative candidates were expressed in bacteria to experimentally validate these proteins as bona fide cAMP-binding proteins. Consequently, we were able to identify new putative effectors of the PKA-independent

pathway in *T. cruzi*.

Identification and characterization of a kinesin, putative BILBO1 partner in the pathogen *Trypanosoma brucei* (P37)

Elodie Berdance, Annelise Sahin, Karen Eguienta, Nicolas Landrein, Denis Dacheux-Deschamps, Mélanie Bonhivers, and Derrick R. Robinson

Laboratoire de Microbiologie Fondamentale et Pathogénicité UMR-CNRS 5234 Bat 3A 1er étage 146 rue Léo Saignat 33076 BORDEAUX Cedex

Trypanosoma brucei brucei causes Nagana. Vaccination is not possible because of antigenic variation and current treatments are difficult to implement or are toxic. For this reasons it is urgent to find new therapeutic targets in order to develop effective treatments. *T. brucei* has a single copy flagellum that emerges from a structure called the Flagellar Pocket (FP), an invagination of the pellicular membrane and the unique site for endo- and exocytosis. The FP is essential. At the neck of the FP there is a cytoskeletal structure: the Flagellar Pocket Collar (FPC) that forms a "ring or "horseshoe" around the flagellum. The FPC consists of numerous proteins, including the first to be identified - BILBO1, which is essential for FP biogenesis. A number of potential BILBO1 partners were identified and are being studied in the lab. Here we characterize one of these proteins: FPC5, a putative kinesin. For this we performed: IFA, RNAi knockdown, over-expression of tag protein in parasites and heterologous expression. FPC5 is cytoskeletal and is localized to the basal bodies. Growth defects are observed after RNAi induction in procyclic and bloodstream forms, and also induces the apparition of zoids, multinucleated cells and detached flagella, which suggests defects in cytokinesis. When expressed in U-2 OS cells, FPC5 does not appear to interact with microtubules as it is normally expected for a kinesin. We are currently purifying the motor domain of FPC5 and will carry out functional analysis *in vitro* for ATPase activity and microtubule co-sedimentation.

Crystal structure and functional inhibition of *Plasmodium falciparum* Thioredoxin Reductase, a validated drug target (P38)

Giovanna Boumis1, Francesco Angelucci3, Andrea Bellelli1,2, Maurizio Brunori1,2, Gianni Desiato1, Serena Pretola1, Fulvio Saccoccia1 and Adriana E. Miele1

1 Department of Biochemical Sciences and Istituto Pasteur – Fondazione Cenci Bolognetti, "Sapienza" University of Rome, 2 CNR Institute of Molecular Pathology and Biology, "Sapienza" University of Rome, 3 Department of Life, Health and Environmental Sciences, University of L'Aquila

Plasmodium falciparum is the vector of the most prevalent and deadly form of malaria, and the search for new drug targets for malaria therapy is urgent, due to the onset of drug resistance. Thioredoxin Reductase (TrxR) is the first step of the NADPH-dependent thiol-mediated detoxification pathway against reactive oxygen species and is essential for the parasite survival during the erythrocytic phase. We solved the structure of recombinant *P. falciparum* TrxR at 2.9Å [1]. The enzyme is an obligate homodimer with three domains: a NADPH binding domain, a FAD-binding domain, and a monomer-monomer interface. The redox activity of the enzyme is accomplished by the electron transfer from the donor NADPH to the FAD cofactor and the first redox active cysteine couple nearby; this reaction is followed by the electron transfer to a second redox center (CGGGKCG) located at the C-terminal arm of the protein that in turn reduces the active disulfide bond of the final substrate, thioredoxin. We assayed the inhibitory activity of different classes of compounds, acting both reversibly or irreversibly on PfTrxR. The gold containing drug Auranofin inhibits TrxR by releasing a gold atom that forms a stable, irreversible complex with one or more cysteine couples, as already observed in the crystal structure of the related enzyme Thioredoxin Glutathione Reductase from *Schistosoma manson*i, solved in our lab. The irreversible inhibition was confirmed in PfTrxR; preliminary crystallization screenings were made, and we are trying to improve the conditions to obtain well diffracting crystals. We assayed the inhibition by two other compounds, 120CF and 556CF, both azoxycyanide derivatives. In both cases the inhibition is apparently reversible and non-competitive. Moreover, for 120CF an apparent Ki in the nM range can be calculated when the activity was assayed with DTNB as substrate. Despite extensive robotic crystallization screening, up to now no crystals were obtained of PfTrxR in complex with reversible inhibitors. This could be due to the complexity of the mechanism of interaction of these compounds with the enzyme, and to the possibility of the molecule to be modified and interact to many possible protein sites.

Estimation of genetic parameters for the resistance to gastro-intestinal nematodes in thoroughbred Arabian horses (P39)

Guillaume Sallé1,2*, Sławomir Kornaś3*, Marta Skalska3, Ingrid David4, Anne Ricard5, Jacques Cabaret1,2

1 INRA, UMR1282 Infectiologie et Santé Publique, F-37380 Nouzilly, France, 2 Université François Rabelais de Tours, UMR1282 Infectiologie et Santé Publique, F-37000 Tours, France, 3 Department of Zoology and Ecology, University of Agriculture of Krakow, 30-059 Krakow, Poland, 4 INRA, UMR1388 Génétique, Physiologie et Systèmes d'élevage, F-31326 Castanet-Tolosan, France, 5 INRA, UMR1313 Génétique Animale et Biologie Intégrative, F-78352 Jouy-en-Josas, France

Equine internal parasites, mostly small Strongyles and *P. equorum*, affect both horses' welfare and performances. The arising of anthelmintic resistant parasites and the need for profitability of the equine industry both urge for optimizing drenching schemes. This optimization may be achieved by identifying some genetic markers associated to susceptibility and to drench carriers of these markers. Our study aimed at characterizing the genetics of horse resistance by estimating heritability of this trait in an Arabian thoroughbred population. A population of 835 Arabian thoroughbred horses from the Michałów stud farm have been measured for *Strongyles* and *P. equorum* infections over 8 years. Analyses showed that 4 and 8% of the observed variation for infection by *Strongyles* and *P. equorum* respectively in this population had a genetic origin. However correction for the imperfect sensitivity and specificity of fecal egg count in horses suggested these estimates would actually be in the range of 15 to 20%. Additional analyses highlighted the dynamics of horse-nematode interaction as fecal egg count in adult horses had a different genetic structure than in zero to five year-old horses. These results suggest a significant part of the inter-individual variation has a genetic origin. The genetic architecture of resistance to nematodes in horses still remains to be addressed but it might provide predictive markers of susceptibility allowing personalised-based drenching schemes.

Resistome of *Leishmania donovani*: multi-factorial genomic origin of clinical antimony resistance (P40)

Jean-Claude Dujardin1, Imamura H1, Mannaert A1, Downing T2, Rijal S3, Sundar S4, Rai K4, Bhattarai N4, Vanaerschot M1, Berg M1, Berriman M5, Cotton J5.
1 Institute of Tropical Medicine, Antwerp, Belgium, 2 NUI, Galway, Ireland, 3 BPKIHS, Dharan,Nepal, 4 BHU, Varanasi, India, 5 WTSI, Hinxton, UK

We sequenced the whole genome of 203 *L. donovani* clinical isolates collected in the last decade in the Indian sub-continent, to characterize the main genomic modifications (resistome) of antimony-resistant (SbR) parasites in an unbiased way. The main population in our sample set was quite homogeneous (2,418 SNPs), but could be split in 9 sub-populations. We focused on one of them (ISC005), which likely emerged 50 years ago (median estimate) and contains most of the SbR isolates from treatment failure. ISC005 parasites share a series of genomic adaptations. First, 32 specific SNPs: 8 nonsyn (among others in a rhomboid serine peptidase), 3 syn, 9 adjacent to a gene. Secondly, a 2-nt indel (mostly homozygous) was observed in the gene coding for aquaglyceroporin, a transporter involved in the uptake of SbIII. Thirdly, two episomes found in all Core-191 isolates showed a significantly higher copy number in ISC005: these amplicons corresponded respectively to a "(de-) phosphorylation kit" and the H-locus involved among others in SbIII efflux. Fourthly, chromosome 22 was significantly more aneuploid than in other populations. Interestingly, we found intermediate SbR parasites appearing to be hybrids between ISC005 and another subpopulation: among others, these parasites showed a clear heterozygosity of the aquaglyceroporin indel. Further work is required to complete the *L. donovani* resistome: especially, (i) characterisation of the kinetoplast genome, (ii) linking these genomic adaptations with the ISC005-specific features encountered by metabolomics and biological studies. We demonstrate here the extent of the molecular adaptations that have accumulated in clinical SbR

Genome profiling of sterol synthesis shows convergent evolution in parasites and guides chemotherapeutic attack (P41)*

Matthias Fuegi1,2, Kapila Gunasekera3, Torsten Ochsenreiter3, Xueli Guan1,2, Markus R. Wenk1,2,4,5, Pascal Maeser1,2
1 Swiss Tropical and Public Health Institute, Basel, Switzerland, 2 University of Basel, Basel, Switzerland , 3 Institute of Cell Biology, University of Bern, Bern, Switzerland, 4 Department of Biochemistry, Yong Loo Lin School of Medicine, National University of Singapore, Singapore, 5 Department of Biological Sciences, Faculty of Science, National University of Singapore, Singapore

Sterols are an essential class of lipids in eukaryotes, where they serve as structural components of membranes and play important roles as signaling molecules. Sterols are also of high pharmacological significance: cholesterol-lowering drugs are blockbusters in human health, inhibitors of ergosterol biosynthesis are widely used as antifungals. Inhibitors of ergosterol synthesis are also being developed for Chagas' disease, caused by *Trypanosoma cruzi*. Here we develop an *in silico* pipeline to globally evaluate sterol metabolism and perform comparative genomics. We generate a library of hidden Markov model-based profiles for 42 sterol biosynthetic enzymes which allows expressing the genomic make-up

of a given species as a numerical vector. Hierarchical clustering of these vectors functionally groups eukaryote proteomes and reveals convergent evolution, in particular metabolic reduction in obligate endoparasites. We experimentally explore sterol metabolism by testing a set of sterol biosynthesis inhibitors against trypanosomatids, *Plasmodium falciparum, Giardia*, and mammalian cells, and by quantifying the expression levels of sterol biosynthetic genes during the different life-stages of *T. cruzi* and *T. brucei*. The phenotypic data correlate with genomic make-up for simvastatin, which showed activity against trypanosomatids. Other findings, such as the activity of terbinafine against *Giardia*, are not in agreement with the genotypic profile.

Glycosylphosphatidylinositols of *Babesia divergens* have unique effects on host cells of the innate immune system (P42)

Françoise Debierre-Grockiego1, Stéphane Delbecq2, Céline Ducourneau1, Isabelle Dimier-Poisson1, Ralph T. Schwarz3, Emmanuel Cornillot2

1 *UMR 1282 Infectious diseases and Public Health, University of Tours - INRA Nouzilly, France, 2 Vaccination Antiparasitaire: Laboratoire de Biologie Cellulaire et Moléculaire EA4558, Université Montpellier 1, France, 3 Institute for Virology, Laboratory of Parasitology, Philipps University, Marburg, Germany*

Babesiosis is a tick-borne disease with a world-wide distribution caused by intraerythrocytic apicomplexan parasites of the genus *Babesia*. Human infection has been mainly associated with *B. microti* and *B. divergens*, with increasing cases of transmission through blood transfusion. In asplenic or immune-compromised patients, infections may cause a malaria-like syndrome and can lead to death. Understanding the molecular basis of host-*Babesia* interactions is a prerequisite for development of initiatives to control babesiosis. It has been shown that glycosylphosphatidylinositols (GPIs) of other parasitic protozoa are responsible for inflammatory effects. The study of *Babesia* GPIs may help us to determine whether they also play an important role in the development of the pathology of babesiosis. *B. divergens* GPIs were individually purified after metabolic labelling and separation by thin layer chromatography. RAW264.7 macrophages incubated with *B. divergens* GPIs differentiated into a M2 phenotype with IL-5 and IL-10 but no TNF-α or nitric oxide production. *B. divergens* GPIs also induced apoptosis of mouse primary macrophages. Expression of major histocompatibility complex class II was decreased at the surface of dendritic cells in response to *B. divergens* GPIs. In addition, GPIs of B. *divergens* prolonged the activated partial thromboplastin time, suggesting a direct effect of GPIs on coagulation factors of the intrinsic pathway. These results suggest that the GPIs of *B. divergens* are unique in their biological effects compared to those induced by GPIs from other protozoan parasites. Characterization of their structures will help us to understand a structure/function relationship.

A class of curcumin analogues with high efficacy against *Trypanosoma* parasites depletes the cells of thiols including glutathione and trypanothione (P43)

Abdulsalam Alkhaldi1,2, Darren Creek2, Hasan Ibrahim2, Michael P. Barrett2,3, Apichart Suksamrarn4, Harry P. de Koning2

1 Department of Biology, College of Science, Aljouf University, Sakaka, Kingdom of Saudi Arabia, 2 Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, UK, 3 Wellcome Trust Centre for Molecular Parasitology, University of Glasgow, Glasgow, UK, 4 Department of Chemistry, Faculty of Science, Ramkhamhaeng University, Bangkok, Thailand.

A class of anti-parasitic curcumin analogues containing a mono-enone linker motif were investigated for their mechanism of action on *Trypanosoma brucei*. The compounds have nanomolar activity against various parasites and low toxicity against human cell lines. Their strong trypanocidal activity is specific to analogues carrying a C7 linker with a C4-C5 double bond and a single ketone group at C3 (enone linker); other curcumin analogues display 1 - 2 orders of magnitude less activity. Mono-enone curcumins including the analogue AS-HK014, did not affect signal transduction in *T. brucei*: no changes were observed in intracellular calcium, cAMP, membrane potential, mitochondrial membrane potential, cell cycle progression or DNA integrity. An AS-HK014-resistant line was produced and found to be resistant only to the mono-enone curcumins, not to the original di-enone curcumin and analogues with similar linker chains. Comparative metabolomics between wild-type and AS-HK014-resistant cells before and after AS-HK014 exposure revealed that exposure of sensitive cells to the analogue (but not to curcumin) leads to the almost complete depletion of intracellular glutathione and trypanothione. An adduct of AS-HK014 and glutathione could be identified in these cells, reproduced by chemical reaction and verified by NMR and mass-spectroscopy. In the resistant line a similar amount of adduct was observed but no significant depletion of thiols occurred in these cells. It is unclear how the resistance mechanism works, as there was no change in the cellular levels of glutathione synthetase (GS), γ-glutamylcysteine synthetase (γGCS) or

glutathione, nor mutations in the GS and γGCS open reading frames.

Purification and characterization of a host-derived chymotrypsin-like enzyme found in adult *Schistosoma mansoni* from infected mice (P44)*

Joseph Egbenya Igetei1,*, Jan Bradley1, Marwa Elfaham1, Susan Liddell2, Mike J. Doenhoff1
1 School of Life Sciences, University of Nottingham, Nottinghamshire, NG7 2RD, UK, 2 School of Biosciences, University of Nottingham, Sutton Bonington Campus, LE12 5RD, UK.

A chymotrypsin-like serine protease found in detergent extracts of *S. mansoni* adult worms perfused from infected mice has been purified and characterized. The enzyme is approximately 85 kDa and hydrolyses NAPBNE (N-acetyl-DL-phenylalanine β-naphthyl–ester), the chromogenic substrate for chymotrypsin-like enzymes. The enzyme appears antigenically and enzymatically similar to a molecule that is present in normal mouse plasma and so is apparently host-derived. The enzyme has been partially purified by depleting normal mouse serum of albumin using sodium chloride and cold ethanol, followed by repeated rounds of purification by one dimensional sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE). After each electrophoresis the part of the gel containing the enzyme was excised, the enzyme eluted from the excised gel segments and concentrated from the eluate. The purified material was analysed by tandem mass spectrometry and identified two peptides; a mouse carboxylesterase 1C (CES1C) and alpha-1B-glycoprotein. The ability of the enzyme to hydrolyze α- or β-naphthyl acetates, which are general esterase substrates, along with other observed esterase characteristics identified it as a carboxylesterase. A similar carboxylesterase has been purified and characterized from rat plasma. Additional evidence to support identification of the enzyme as a carboxylesterase, and possible roles of the enzyme in the host-parasite relationship, will be discussed.

Analysis of force of infection trends for Chagas disease in Colombia (P45)*

Zulma Cucunubá1,2*, Pierre Nouvellet1 , Víctor M. Angulo2, Gabriel J. Parra2, María-Gloria Basáñez1
1 Department of Infectious Disease Epidemiology, Imperial College London, Norfolk Place, London W2 1PG, UK, 2 Red Chagas Colombia, Instituto Nacional de Salud. Av-calle 26 51-20, Bogotá, Colombia, 3 MRC Centre for Outbreak Analysis and Modelling, Imperial College London, Norfolk Place, London SW2 1PG, UK, 4 Centre for Health Policy, Imperial College London, South Kensington, 1st Floor Sherfield Building, London, SW7 2AZ, UK

Chagas disease prevalence estimates are insufficient to understand the dynamics of this disease in human populations, especially when time of exposure is not considered. This study aimed to analyse force of infection (FOI) trends for Chagas disease in Colombia using sero-prevalence data from different locations. We used two catalytic models: a simple model considering a constant FOI as a function of exposure time (where age structure accounts for exposure time), and a transmission interruption model that includes a change (reduction) of FOI at a specific (to be estimated) time. Parameters were estimated by maximum likelihood using R. Overall, 54,758 registries for 14 departments were obtained. Using the simple model, in Casanare the FOI was estimated at 0.01 person/year (95%CI: 0.009-0.012) from a survey conducted in 2000 (which accounts for the exposure period 1985-2000), and as 0.003 (95%CI: 0.002-0.005) in a 2009 survey (period 1991-2009). In Boyacá, the FOI was 0.08 (95%CI:0.07-0.09) in a 1995 survey (period 1920-1995) and 0.001 in a 2010 survey (period 1967-2000). In Santander, the FOI was 0.15 (95%CI: 0.13-0.17) in 1998 (period 1919-1998), and 0.013 (95%CI: 0.01-0.017) in 2011 (period 1968-2011). Using the interruption model, a substantial reduction in the FOI is suggested for Casanare around the 1970's. These results suggest a decrease of the incidence trends in some locations. Further analyses are needed to explore whether these changes have resulted from implemented control strategies, housing improvements or migration movements. Uses and limitations of this approach will be discussed.

Composition of the gut Microbiota in the naturally parasitised yellow-necked mouse, *Apodemus flavicollis* (P46)

Geraldine Bastien1, Jakub Kreisinger1, Emily Pascoe1, Heidi Hauffe1 and Sarah E. Perkins1,2
1 Department of Molecular Ecology, Foundation Edmund Mach, San Michele all'Adige, Italy, 2 The Sir Martin Evans Building, Museum Avenue, Cardiff University, Cardiff, United Kingdom, CF10 3AX

The gut microbiota plays a crucial role in vital host functions, such as brain and immune system development. Emerging evidence indicates that parasitic helminths also interact with the gut microbiota to affect host functions. However, to date, the interaction between host, microbiota and helminths is not well studied. Wild rodents, such as the yellow-necked mouse, are naturally parasitized, and thus represent a promising model to start to address this question. As such, we have investigated the variability of the gut microbiota in five distinct locations of the gut, where parasites are

commonly found. We analysed the variability of the gut microbial community in the small intestine, caecum, proximal and distal colon within fifteen yellow-necked mouse *Apodemus flavicollis*. A higher proportion of reads were assigned to the order *Bacteroidales* and *Clostridiales* in faecal samples and the caecum, whereas the small intestine and intestinal membrane were dominated by *Lactobacilliales*. These results at the class and order level are consistent with previous analyses of other mammal's gastrointestinal microbiota. Most interestingly, our results at a lower taxonomic level suggest considerable variation in the composition of the microbiota colonizing the different parts of the gut. Further analyses will determine to what extent this result is related to the presence of parasitic helminths at these locations. To our knowledge this study is the first providing insight into compartment-dependent variation of gastrointestinal microbiota composition in a wild rodent non-captive population.

Characterisation of a glycosylated glutathione transferase of *Onchocerca ochengi* (P47)

James LaCourse1*, Acosta-Serrano, Alvaro1, Makepeace, Ben2, Davis, Jem1,Molyneux, Gemma1, Armstrong, Stuart2, Perally, Samirah1, Rutter, Anne1, Prescott, Mark2
1 Liverpool school of Tropical Medicine, 2 University of Liverpool

The cattle filarial nematode *Onchocerca ochengi* is a well-established model for the study of human onchocerciasis, the causative agent of which is *O. volvulus*. Our collaboration involving groups in the University of Liverpool and a field site in Cameroon has recently identified a glycosylated glutathione transferase (GST) from *O. ochengi*, homologous to an immunodominant and potential vaccine candidate from the human parasite *O. volvulus*. The screening of parasite products for immune reactive vaccine candidate antigens and subsequent identification is predominantly protein focused. However, a comparison of antibody responses to eukaryotic- and prokaryotic-expressed recombinant proteins and/or glycosylated or deglycosylated proteins demonstrates the high immunogenicity of N-glycans extensions. Additionally, the potential role of glycans in active secretion of this enzyme to function in, as yet undefined, roles at the host-parasite interface is also of particular interest. Therefore, an understanding of carbohydrate modifications of proteins has important implications for future vaccine developments and an understanding of host-parasite interactions. Proteomic and enzymic investigations to isolate, characterise and unravel the structure of N-glycans modifications on this protein and how this may relate to its enzyme activity, immunogenicity and host-parasite interactions are revealed in this study.

S. mansoni* therapeutic effect on DSS-induced colitis through FoxP3+ Tregs and TH1/Th2 paradigm (P48)

Marwa HasbySaad, Eiman A. Saad, Kholoud A. El-Nouby
Tanta University, Faculty of Medicine, Medical Parasitology Department, Egypt

Schistosoma mansoni manipulates the immune response during the sequential phases of infection by shifting between the two T helper cells and stimulation of FoxP3+ Tregs. At the same time, Th cell dysregulation is the main pathological feature of inflammatory bowel diseases. We aimed to investigate the impact of *Schistosoma mansoni* infection phases and antigenic material on DSS-induced colitis (an experimental model for IBDs in mice), and determine Th1/Th2 paradigm and FoxP3+ Tregs role. Mice were exposed to DSS course after acute/chronic schistosomiasis, or after worm homogenate/killed eggs injection. Clinical disease activity index, serum Th1/Th2 cytokines (IFN-γ, IL-2, IL-4 & IL-10), inflammatory scores and FoxP3+ Tregs in colon were measured. We found significant less DAI and macroscopic inflammatory score of colon with chronic infection or antigenic material injection. After DSS, colon showed milder cryptitis with chronic schistosomiasis, moderate with homogenate and marked improvement after killed eggs. IL-4 showed the least level after killed eggs. A significant positive correlation between: IL-4 and IL-10 in groups with DSS and schistosomiasis, and between FoxP3+ Tregs and IL-10 while a significant negative correlation between IL-10 and IL-4 after killed eggs was found. A significant infiltration with FoxP3+ Tregs with *Schistosoma* element, the highest in chronic infection and killed eggs, showed no significant difference. We conclude that *S. mansoni* improved colitis mainly after chronic infection or killed eggs, and that the Th1/Th2 paradigm is the core of helminthic therapy in infection, with a shift to Tregs regulation.

Potential role of microRNAs in host-parasite interaction (P49)*

Henry Gu1, Alan Winter1, Tom McNeilly2, Collette Britton1, Eileen Devaney1
1 *Institute of Biodiversity, Animal Health and Comparative Medicine, University of Glasgow, 2 Moredun Research Institute, Edinburgh*

miRNAs are short 22 nucleotide RNA molecules that regulate gene expression post-transcriptionally. We are investigating possible interaction of parasite miRNAs and the host immune response during *Haemonchus contortus* infection. *H. contortus* is a gastrointestinal nematode and a significant cause of welfare and economic problems in small ruminants. A previous study identified 192 miRNAs in *H. contortus*, one of which, mir-5352, is the focus of this study. mir-5352 is one of a cluster of four miRNAs, which are conserved only in nematodes infecting the gastrointestinal tract. Microarray and qRT-PCR showed that mir-5352 and other members of the cluster are highly expressed only in parasitic stages. Further qRT-PCR experiments identified mir-5352 in excretory-secretory (ES) products of adult parasites. Sequencing of a small RNA library prepared from adult ES material identified mir-5352 as well as 52 other microRNAs in the ES, some of which appear to be selectively released. Work is underway to determine if parasite specific microRNAs can be detected in sheep abomasal tissue and lymph nodes, suggesting a potential interaction with host cells. Bioinformatic prediction programmes identified CD69, an early marker of T-cell activation, as a potential target of mir-5352. A dual luciferase assay utilising the 3' UTR of CD69 is being used to determine if mir-5352 is able to regulate CD69 expression. Combined, these different approaches will give us a greater insight into the potential role of parasite microRNAs within the host.

Extracellular microvesicles in the parasitic nematode *Teladorsagia circumcincta* (P50)*

Thomas Tzelos, Jacqui Matthews, Amy Buck, David Frew, Neil Inglis, Alasdair Nisbet, Bruce Whitelaw, David Knox, Tom McNeilly
Moredun Research Institute

T. circumcincta is a major cause of ovine parasitic gastroenteritis in temperate climatic regions.
The development of anthelmintic resistance by the parasite challenges its future control. Recent research indicates that many parasite species release extracellular microvesicles (EMV) into their environment. These vesicles are considered to play an important role in the intercellular communication between parasites and their hosts, and thus represent potentially useful vaccine and/or drug targets. Here, we wanted to examine whether *T. circumcincta* produces EMV and if so, what proteins they contain. EMV were purified from the ES products of *T. circumcincta*'s fourth larval stage. Transmission electron microscopy (TEM) and proteomic analysis were used to confirm the presence of the vesicles and to identify the proteins they contain, respectively. The TEM confirmed the presence of EMV in the parasites' ES products and proteomic analysis revealed several types of proteins within the vesicles including: Activation-associated secreted proteins, Actins, Metallopeptidases, and RAB proteins. A comparative analysis of EMV, total ES products and ES after the extracellular vesicles purification (SN) showed that approximately 35% of the proteins found in the vesicles could also be identified in total ES and in SN, whilst the remaining 65% were present only in EMV. This study represents the first report, to our knowledge, of EMV production by any ruminant nematode species, and shows that *T. circumcincta* EMV contain unique proteins which could be targeted in future vaccine and/or drug interventions aimed at controlling this important parasite.

Inhibitors of Kinetoplastid Sphingolipid Synthases as Novel Anti-Leishmanial Agents (P51)*

Jennifer Norcliffe1, E. Alvarez-Ruiz2, S. Gonzalez-Del Valle2, J. J. Martin-Plaza2, P. G. Steel1,P. W. Denny1
1 Biophysical Science Institute, Durham University, Durham DH1 3LE, 2 Glaxo Smith Kline, Medicines Development Campus, 28760 - Tres Cantos (Madrid), Spain

The protozoan kinetoplastid parasites *Leishmania* spp*, Trypanosoma brucei* and *Trypanosoma cruzi* are responsible for potentially fatal diseases that affect over 22 million people worldwide, with an estimated 450 million at risk. Current therapies are expensive and not widely accessible. In addition, drug toxicity and emerging resistance are major concerns. Previous work has identified the essential kinetoplastid sphingolipid synthase (SLS) as an attractive pharmaceutical target due to the divergence of function compared with the mammalian orthologue. A high-throughput compatible screening assay was developed to test the 1.8 million compound library held at Glaxo Smith Kline in Tres Cantos against the *Leishmania major* enzyme. The 19,669 compounds identified were subjected to further screening to identify those which were highly active yet selective for the parasite enzyme. 216 of these compounds were selected as being promising drug candidates and were subsequently tested on *Leishmania* promastigotes and amastigotes (the insect and mammalian stages of the parasite respectively), infected macrophages and HepG2 cells (a human liver cell line). The results were encouraging, with several compounds displaying high activity against the parasites whilst having a negligible effect on the human cells. A set of promising chemical scaffolds has subsequently been identified and are presently being evaluated in murine models. Current and future work centres around redesign and resynthesis of these compounds in order to produce the best possible candidates for clinical trials.

A family of heterogeneous telomerically-encoded *Cryptosporidium* glycoproteins (HTEGs) displaying host specific sequence variation (P52)*

Johanna Nader1, Maha Bouzid1, Kristen Elwin2, Rachel Chalmers2, Paul Hunter1, Kevin Tyler1
1 Department of Medicine, Norwich Medical School, University of East Anglia, 2 Cryptosporidium Reference Unit, Public Health Wales, Dept of Microbiology, Singleton Hospital

Genetic variability is limited between the two major human-infective species of Cryptosporidiosis, *Cryptosporidium parvum* and *Cryptosporidium hominis*. A fact which makes it difficult to identify the genetic basis of differential host specificity and virulence. We have previously characterized two proteins, encoded telomerically which display characteristics commonly observed in genes involved in host specificity and virulence. Here we describe the further characterization of an extended HTEG group. Four new members of the HTEG family were identified in silico, including a telomeric gene at the end of chromosome 6 in *C. parvum*, its ortholog in *C. hominis*, a telomeric gene at the end of chromosome 5 of *C. hominis* and its ortholog, which appears to be internal, on chromosome 5 of *C. parvum*. Initial screening across a wide range of cryptosporidium species with an internal portion of this gene amplified most Cryptosporidium strains and species but showed considerable divergence between zoonotic forms of *C. parvum* and *C. baileyi* compared to anthroponotic *C. parvum* and *C. hominis*. Protein divergence was inferred by calculating the ratio of non-synonymous to synonymous SNPs. Analysis of the divergence between zoonotic and anthroponotic reference strains of Cryptosporidium provided a preliminary indication that these genes may be involved in adaptation to the human host. Inclusion of further zoonotic and anthroponotic strains will test this link. The development of these genes for improved species specific assays in diagnosis and risk assessment is described.

Comparative study of glycosylphosphatidylinositols isolated from trypomastigotes and amastigotes of *Trypanosoma cruzi*: opposite biological effects. (P53)

Françoise Debierre-Grockiego1, Philipp Stahl2, Ralph T. Schwarz2, Rudolf Geyer3, Peter Kaese3, Terry Smith4, Céline Ducournau1, Isabelle Dimier-Poisson1
1 *UMR1282 Infectious diseases and Public Health, University of Tours - INRA Nouzilly, France, 2 Institute for Virology, Laboratory of Parasitology, Philipps University, Marburg, Germany, 3 Institute of Biochemistry, University of Giessen, Germany, 4 Biomedical Sciences Research Centre, The North Haugh, The University, St. Andrews, Scotland, UK*

Trypanosoma cruzi, the causative agent of the Chagas' disease that affects eight million people in Latin America, is transmitted by blood-sucking insects, blood transfusion, organ transplantation or congenitally. The parasite persists in tissues for decades causing cardiomyopathy or megasyndromes, and even death. Even though parts of the host-parasite interactions have been elucidated, many interactions remain undefined. It has been shown that glycosylphosphatidylinositols (GPIs) purified from surface mucins of *T. cruzi* trypomastigotes induce the secretion of cytokines in macrophages. In this work we have compared the GPIs purified from trypomastigotes and from intracellular amastigotes. After metabolic labelling and separation by thin mayer chromatography, different profiles of GPI species have been distinguished between the two forms. Individual GPI species (16 for trypomastigotes and 14 for amastigotes) have been extracted, and their glycan and lipid contents showed differences by biochemical analysis. Furthermore, GPIs of the trypomastigotes and amastigotes had opposite biological effects on macrophages and dendritic cells (differences in production of cytokines, surface expression of major histocompatibility complex I and II molecules, and cell death). Free GPIs were detected in the supernatant of *in vitro* culture of amastigotes, suggesting that the GPIs of the intracellular form are able to active cells *in vivo*. It has been reported in the literature that extracellular amastigotes often appeared during *in vitro* culture. Our study indicates that this third form had 16 main GPI species distinct from those of trypomastigotes and intracellular amastigotes.

Malaria: Epidemiology, Diagnosis, Treatment and Control (P54)

Khurram Goraya1, Abdul Hannan2
1 *University of Health Sciences Lahore 54600, Pakistan, 2 University of Health Sciences Lahore 54600, Pakistan*

Malaria is a disease caused by parasites of the *Plasmodium* genus and leads to high morbidity and mortality. Malaria is known as a disease of poor man but, in fact, poverty comes due to malaria. The expenditure of huge money for the treatment of malaria in endemic countries is major cause of their lagging behind the modern world. Being a developing country, malaria is a significant public health problem in Pakistan. Pakistan is among the top 10 countries estimated for the highest population at risk of malaria. Extensive agricultural practices coupled with vast irrigation network, monsoon

rains, large population movement, complex political situation in certain border areas, and high level resistance to antimalarial drugs and insecticides play pivotal role in the spread of malaria. Poor access of the population to early diagnosis, effective treatment and effective prevention measures have further compounded the situation. This article details the biology of *Plasmodium*, diagnosis, treatment and possible indigenous, economical and ecofriendly ways to control malaria.

Ecological dynamics of host/parasite co-distribution: Toxoplasma in a natural rodent population (P55)*

Jaroslav Bajnok, Kellyanne Boyce, Geoff Hide
University of Salford School of Environment and Life Sciences Peel Building Salford M5 4WT

Wood mice (*Apodemus sylvaticus*) were collected from four ecologically distinct locations within a $2km^2$ wooded rural ecosystems with a very low frequency of cats. DNA was successfully isolated from 126 *Apodemus* brains and tested by PCR for the presence of *T. gondii*. Forty four samples gave positive reactions with four *T. gondii* specific markers SAG1, SAG2, SAG3 and GRA6 giving an infection rate of 34.92% (95% CI: 27.14%-43.59%). Detailed analysis of RFLP patterns the SAG2, SAG3 and GRA6 loci showed that 5 had Type II patterns at all three tested loci, 23 had the combination of Type II and Type III alleles and 16 had all three genotypes present. The RFLP patterns were consistent with a high frequency of mixed strain infections. Population genetic analysis of the mice showed that the site consists of three genetically distinct populations spread over the four sampling locations. No significant difference was found in prevalence in males and females ($\chi 2$ =0.863, D.F. = 1, P = 0.353). There was no significant age prevalence effect with *T. gondii* infection (P = 0.23). There was a significant difference in *T. gondii* prevalence between each genetic population ($\chi 2$ =7.950, D.F. = 2, P = 0.018) but not between sampling locations ($\chi 2$ =8.06, D.F. = 3, P = 0.29). Our data showed that the prevalence of *T. gondii* in this natural population is high and the parasite can be perpetuated in the absence of felids.

Seasons of disease: Using baboons in a seasonal environment to predict changes in disease risk due to climate change (P56)*

Cassandra Raby
University of Liverpool; Institute of Zoology Institute of Integrative Biology, Biosciences Building, University of Liverpool, Crown Street, Liverpool L69 7ZB

Seasonal changes provide a source of strong external variation in natural systems. Whilst associations between seasons and infectious diseases have been well documented, it has proven more difficult to determine exactly how and why climatic changes influence disease fluctuations. The aims of this research are to (1) identify the mechanistic links between climate and disease in a model host-parasite system in the seasonal environment of Namibia, Africa, and (2) explore what the consequences of different climate change scenarios might be for disease transmission. The research involves observations of gut parasites in two populations of wild chacma baboons (*Papio ursinus*). This includes two nematode species (*Streptopharagus pigmentatus* and *Physaloptera caucasia*) that have indirect life-cycles involving arthropod intermediate hosts; and protist species (Ciliated protozoan *Balantidium coli*; amoeba commensals and parasites, e.g. *Entamoeba coli* and *Entamoeba histolytica*) that are transmitted directly. Due to the different transmission routes of these parasites we predict that climate (and climate change) will influence their transmission and prevalence differently. The information collected from studying these populations will be used to inform an individual-based model, which incorporates behavioural variation within the population, to predict the long-term changes in disease risk due to climate change. Here I describe the steps I am following to conduct this research, and present preliminary data on the climate-related behaviour patterns of these baboons, and the likely consequences for infection risk in their seasonal environment.

Large-scale growth of *P. falciparum* mature gametocytes in Albumax using a bioreactor. (P57)*

Sonia Lozano, María Jesús Almela, Carolina Gonzalez, Janneth Rodrigues, Esperanza Herreros
Diseases of the Developing World, Glaxo Smith Kline, 28760 Tres Cantos, (Madrid), Spain

The tropical disease malaria caused by *Plasmodium falciparum* is one of the most important infectious diseases which results in more than one million deaths annually and is transmitted by the bite of the Anopheles mosquitoes. Parasite transition from the human host to the mosquito vector is mediated by gametocytes, sexual stages that are formed in human erythrocytes, which play a crucial part in the transmission of the disease. The biology of gametocytes is complex and consists of five morphologically identifiable stages: stages I to IV (immature gametocytes) sequestered into human

tissues; and stage V (mature gametocytes) which circulate in the blood stream. For the studying the effect of anti-malarial's on the sexual stages of the parasite is to effectively produce large quantities of mature stage V gametocytes *in vitro*. Production of large amounts of gametocytes using current techniques requires time consuming and labor intensive methodologies which involve several small flasks with daily media changes. Presently there are very few methods reported for large-scale production of gametocytes. We report here the development of a system for scale-up of gametocyte production using a bioreactor device and culture medium with Albumax with 3D7A *P. falciparum* strain. The method we describe presents several major advantages over the static flask method like non-requirement for regular media changes. This method is less time consuming, provides us with more reproducible results and large amounts of viable and robust Stage V gametocytes for screening anti-malarial's with transmission blocking potential in an industrial scale HTS platform.

Characterisation of the moulting process in the salmon louse, Lepeophtheirus salmonis, and its potential as a vaccine target (P58)

Carol McNair, Sean Monaghan, Qiwen Zhong and James Bron.
Institute of Aquaculture Pathfoot Building University of Stirling Stirling FK9 4LA

Sea lice are a major pest of both farmed and wild Atlantic salmon (*Salmo salar*) worldwide. The most important species found in the UK is *Lepeophteirus salmonis*. Sea lice cost the Scottish salmon industry £33 million every year, making them a major concern in Aquaculture. Treatment of lice is usually through the use of chemotherapeutants, but resistance to these is rapidly increasing. Hence alternative control methods are being investigated. One possible alternative would be the development of an anti-louse vaccine. For a vaccine against *L. salmonis* to be effective, it would have to target an antigen which was either essential to the louse's survival, feeding or development. The moulting process (ecdysis) could present a novel target for vaccination. Sea lice attach to their host at the copepodid stage, and proceed to moult to chalimus stages, then pre-adults and eventually adults. If this moulting process could be interrupted, the lice would not be able to progress to the damaging feeding stages. Ecdysis is not yet well understood in crustaceans, however the process is well-documented in other arthropods such as *Drosophila melanogaster*. By analogy to *D. melanogaster,* homologues of enzymes involved in the production of moulting hormone (ecdysone) have been identified in *L. salmonis*. These homologues have been amplified from different lifecycle stages of lice and localised using in situ hybridisation. Studies are ongoing to ascertain whether targeting these enzymes would interrupt moulting in the louse, and thus prevent damage to the host.

Different hemozoin components promote chemokines production by human microvascular endothelial cells (P59)

Nicoletta Basilico1, Yolanda Corbett2, Sarah D'Alessandro2, Silvia Parapini2, Mauro Prato3, Paolo Arese4, Donatella Taramelli2
1 Dipartimento di Scienze Biomediche, Chirurgiche e Odontoiatriche, Università di Milano, Milan, Italy, 2 Dipartimento di Scienze Farmacologiche e Biomolecolari, Università di Milano, Milan, Italy, 3 Dipartimento di Neuroscienze and Dipartimento di Scienze della Sanità Pubblica e Pediatriche Università di Torino, Turin, Italy, 4 Università di Torino, Turin, Italy

Cerebral malaria (CM) is a fatal complication of *Plasmodium falciparum* infection caused by the cytoadherence of infected erythrocytes to brain endothelial cells followed by micro-circulatory obstruction, blood-brain barrier (BBB) damage, inflammatory response and neurological sequelae. The combination of both parasite and host factors causes the pathogenesis of CM. In particular, malarial pigment, hemozoin (HZ) has been shown to interfere with monocytes and endothelial cell functions. HZ is a ferriprotoporphyrin-IX crystal bound to host and parasite proteins and lipids. HZ is able to generate lipo-peroxidation products such as 15(S,R)-hydroxy-6,8,11,13-eicosatetraenoic acid (15-HETE) and 4-hydroxynonenal (4-HNE) and it seems to be constantly associated to host fibrinogen. In the present work human dermal microvascular endothelial cells (HMEC-1) were treated with the full native HZ, isolated from *P. falciparum* cultures, and with specific HZ components (fibrinogen, 15-HETE, 4HNE, ferriprotoporphyrin-IX crystals). Chemokines (CXCL-8, CCL-5) production was evaluated by ELISA. HZ stimulated the production of the chemokines CXCL-8 and CCL-5 in a dose and time-dependent manner, however when delipidized HZ or lipid-free synthetic HZ (beta-haematin) were used, no significant changes in the chemokines release were observed. On the contrary, both fibrinogen and 15-HETE, but not 4-HNE, were able to induce significant amount of chemokines. The present data suggest that chemokines release from HZ-stimulated endothelial cells could be due to different and specific hemozoin components. As a consequence of chemokines production, leucocytes recruitment is favoured resulting in the amplification of the inflammatory response and concurring to BBB impairment in CM.

Impaired CXCR4/CXCL12 chemokine receptor/ligand axis limits filarial infection (P60)

Nicolas Pionnier1,2, Emilie Brotin2, Gregory Karadjian1, Nathaly Vallarino-Lhermitte1, Adélaïde Nieguitsila1 Patrice Hemon3 Marie-Laure Aknin2, Françoise Bachelerie2, Coralie Martin1,
1 UMR 7245 MCAM MNHN-CNRS Muséum National d'Histoire Naturelle 61 rue Buffon CP52 75005 Paris France, 2 UMR-S996 INSERM - LabEx LERMIT Cytokines, Chemokines and Immunopathology 32 rue des Carnets 92140 Clamart France, 3Plateforme Histologie PHIC UnivSud IFR-141 IPSIT Faculté de Pharmacie 92296 Châtenay-Malabry France

Filariases are chronic diseases affecting 160 million people worldwide. Despite considerable effort to reduce disease burden, particularly through mass drug administration programs, filarial infections remain a major public health problem requiring new therapeutic approaches. In our study, we used *Litomosoides sigmodontis* as a well-established murine model of filarial infections. Previous studies have shown that the CXCL12 chemokine and its receptor CXCR4 participate to the mice resistance mechanism to the filarial infection, suggesting CXCR4 and CXCL12 as potential therapeutic targets. To decipher their role on the infection progression, we used a newly developed murine model we developed of a rare combined human immunodeficiency disorder (WHIM: Warts, Hypogammaglobulinemia, recurrent Infections and Myelokathexis) caused by a gain of CXCR4 function and also characterised by a profound lympho-neutropenia. WHIM mice reproduce this leucopenia, associated with defective thymopoiesis and B-cell development and lymph node disorganised architecture. Our results on filarial infection in those mice showed that filarial parasitic success was decreased by 70% in WHIM mice compared to control wild-type mice. In addition, a significant neutrophilia became noticeable from 15 days post-infection, normalising the circulating neutrophils to the control levels although the lymphopenia remained in the WHIM mice throughout the infection. Moreover, before filarial injection, cell recruitment in the skin was more important in WHIM mice, suggesting a more efficient local immune response to *L. sigmodontis* thereafter. Further analyses are currently conducted in order to elucidate the mechanisms behind this immune response in respect to the role of the CXCL12/CXCR4 axis in filarial infection.

Understanding the molecular mechanisms of refractoriness to a trypanosome infection in *Glossina morsitans morsitans* (P61)*

Nicholas Ejeh, Stella Lehane ,Lee R Haines, Neil Hall, Michael Lehane, Alistair Darby, Alvaro Acosta-Serrano
Liverpool School of Tropical Medicine

Tsetse flies are naturally resistant to trypanosome infection with only ~0.1% of wild flies carrying mature infectious forms. The mechanisms behind tsetse refractoriness are not well understood at the molecular level, but the identification of tsetse molecules affecting establishment of a trypanosome infection in the fly are essential for the design of future vector control strategies. The tsetse refractory phenotype observed in the field can be reproduced in the lab by feeding flies with normal blood prior to receiving an infected bloodmeal. We used these conditions to produce refractory *Glossina morsitans* flies. Midguts from refractory flies were dissected several days post-infection with *Trypanosoma brucei* bloodstream forms to generate a transcriptome library for 454 pyrosequencing. We found that a total of 184 genes were differentially expressed (DE) from which 70 were selected based on those that met the criteria for down-regulation and up-regulation (i.e. 1.2 and above for up regulated genes and -1.15 and below for down-regulated genes). Both the up- and down-regulated genes were involved in various pathways such as immune regulation, metabolism, cell death, cellular and cell signalling. Out of the 70 DE candidate genes, 17 were significantly up-regulated. RNA interference (RNAi) was then used to validate the functions of three up-regulated candidate genes. RNAi ablation of each of these individual genes was able to revert the refractoriness phenotype up to 50%, with knockdown levels of ~70% as determined by qPCR. We are currently determining the role of these candidate genes during a trypanosome infection

Evidence of *Toxoplasma gondii* in a Scottish catchment (P62)*

Hannah Shaw, Beth Wells, Elisabeth Innes, Frank Katzer
Moredun Research Institute, Edinburgh, EH26 0PZ

Waterborne transmission of *Toxoplasma gondii* is increasingly been considered as a public health threat due to documented incidences of contaminated drinking water, leading to serious outbreaks in the human populations. A study was conducted to optimise the method for extracting *T. gondii* oocysts from water, and also to use this method to

determine the *T. gondii* load in water samples from a Scottish catchment where *T. gondii* was detected in water previously. Currently, the standard method for extracting *Toxoplasma gondii* oocysts from water samples is cartridge filtration, followed by sucrose flotation and centrifugation before undergoing DNA extraction and real time PCR to detect the 529bp repeat element present in the *T. gondii* DNA. The method was optimised by using the Macherey-Nagal NucleoSpin DNA Extraction Kit along with the addition of a flotation step for dirty water samples. 36 water samples were obtained for the catchment in January and February 2014. These samples were processed using the adapted technique mentioned above. *T. gondii* DNA was found in 8 of the 36 water samples showing that the adapted technique is successful in extracting oocyst DNA from the water samples. This data was compared with rainfall data obtained from the MET office in order to correlate *T. gondii* oocyst load with rainfall events. It was found that the oocyst load increased during a high rainfall event.

Feline patent toxoplasmosis among feral cats (*Felis catus*) in Doha city, Qatar and its immediate surroundings (P63)
Marawan Abumadi, Marawan Abu-Madi
Qatar

Doha city has a high feral cat population and studies of hospital records in Doha have shown that human toxoplasmosis also occurs. In this study we aimed to assess the extent of patent toxoplasmosis among feral cats sampled between June 2008 and April 2010, from a range of locations radiating out of the city centre in concentric semi circular/elliptic rings and by north, west and south divisions within each of the rings. A total of 4,652 cats were sampled and overall prevalence of infection was 9.1%. Prevalence was 10.1% in the first summer, and then dropped to 8.4% in the following winter and further to 6.8% in the next summer before rising to 10.6% in the final winter of the study; this interaction between annual period and season was significant. There were also significant changes in prevalence across each of the consecutive months of the study, but no clear pattern was evident. Prevalence did not vary significantly by city sector and there was no difference in prevalence between the host sexes. We conclude that despite minor, and significant perturbations, the prevalence of patent *T. gondii* infections among cats in Doha is remarkably stable throughout the year, across years and spatially within the city's districts.

A molecular approach to identification of nematodes from harbour porpoises (*Phocoena phocoena*) (P64)
Ruth Kirk[1], Rebecca Boulert[1], Sophia Girgis[1], Paul D. Jepson[2], Ryan Lee[1] and Scott P. Lawton[1]
[1]School of Life Sciences, Kingston University, Surrey KT1 2EE, UK, [2]Institute of Zoology, Zoological Society of London, Regent's Park, London NW1 4RY, UK

The common harbour porpoise is a highly mobile, small cetacean that lives in the North Atlantic and North Pacific Oceans and in some semi-enclosed seas. Their coastal distribution makes them highly susceptible to anthropogenic pressures such as fishing, acoustic and chemical pollution, so they are listed as endangered in several international conservation instruments. The UK Cetacean Strandings Investigation Programme (CSIP) has reported that one of the principal causes of death in UK-stranded harbour porpoises were infectious diseases, mainly pneumonias due to a combination of parasitic, bacterial and fungal infections. It is therefore important to continue to monitor the incidence of disease in stranded harbour porpoises in order to identify any new threats to their conservation status. The present study was undertaken to evaluate a molecular approach to identifying nematodes which had previously been identified using morphology. Nematodes were removed during post-mortem examinations of stranded or by-caught harbour porpoises. DNA barcoding methods were applied using ribosomal ITS-2 regions. The porpoises were infected with *Metastrongyloidea* lungworms such as *Pseudalius inflexus, Stenurus minor* and *Torynurus convolutus*, and *Anisakis* stomach worms. Phylogenetic relationships amongst lungworm species and the feasibility of using DNA barcoding techniques as a routine method of identification are discussed.

Toxoplasmosis: Revisiting Prevalence, Transmission, Genotyping, Treatment and Control (P65)*
Saher Islam1*, Amna Arshad Bajwa1, Wasim Shehzad1, Kamran Ashraf2, Imran Rashid2, Haroon Akbar2
1 Institue of Biochemistry and Biotechnology, University of Veterinary and Animal Sciences, Lahore, Pakistan, 2 Department of Parasitology, University of Veterinary and Animal Sciences, Lahore, Pakistan

Toxoplasma gondii, a well known intracellular parasite from phylum apicomplexa, causes toxoplasmosis in almost all warm blooded animals around the globe. About one-third human population has been infected by this parasite. It causes severe risks and economic losses to public health as well as to livestock industry. The protozoan has a heteroxenous life

cycle and cats act as definite host. The protozoa can be transmitted horizontally as well as vertically. It is now the subject of renewed interest, due to its several routes of transmission causing severe toxoplasma infections among diverse hosts. This infection may be acute, chronic and asymptomatic depend on immune status of an individual. As this protozoan has one species *T. gondii*, but has various strains. Virulence of *T. gondii* infection varies among animals, depending on its strain. Genotype of infected strains has great impact on clinical results of toxoplasmosis. It helps to improve our understanding towards the type of the infection associated with genotype, possible outcomes of the disease and its treatment. We highlight the importance of different genotyping methods to understand the population genetics each with their advantages and limitations. The disease can be diagnosed applying various methods, from microscopy to serological examination. Advancements in molecular techniques have revealed new series of molecular markers and PCR-based approaches are helpful to understand the genetic diversity among strains. Further, it provides important basis for diagnosis and treatment. Different strategies from drug therapy to vaccination and control measures are being adopted to reduce the risk of infection.

Evolution of functional diversity in the trichostrongylid levamisole receptor (P66)

Robin Beech1,2, Thomas Duguet1, Aynsley Merk1, Claudia Wever1, Joe Dent1, Claude Charvet2, Cedric Neveu2
1 Institute of Parasitology, McGill University, Canada, 2 ISP, INRA, Tours, France

Levamisole is widely used to cure parasitic nematode infections, where it binds to and activates a class of acetylcholine receptors (L-AChRs) expressed at neuromuscular junctions. The L-AChR of the model nematode *Caenorhabditis elegans* contains subunits encoded by unc-38, unc-63, lev-8, lev-1 and unc-29. In contrast, *Haemonchus contortus* possesses four copies of unc-29 and the L-AChR contains in addition, unc-38, unc-63 and acr-8. UNC-29.1, UNC-29.3 and UNC-29.4, but not UNC-29.2 produce functional channels in Xenopus oocytes when combined with *H. contortus* subunits, with similar affinity for ACh but differing response to LEV and other ligands. All four are functional when expressed with the remaining subunits from *C. elegans* in Xenopus. This is confirmed *in vivo* by rescue of a *C. elegans* unc-29 KO by transfection of each unc-29 copy. This suggests co-adaptation between UNC-29.2 and the other H. contortus subunits is responsible for the lack of a receptor, rather than changes limited to UNC-29.2. Specific UNC-29 copy antibodies allow us to localize the novel L-AChR targets in *H. contortus in vivo*. The UNC-29 copies of *H. contortus* are functionally divergent and produce receptors with distinct pharmacology. Comparison of the different subunits and the unusual properties of UNC-29.2 provides an experimental platform to investigate the details of ligand binding, gating and the evolution of subunit assembly. Taken as a whole, this work establishes, critically, that evolutionary differences between nematode species must be taken into account when using *C. elegans* as a model and that parasite receptors must be examined directly.

Towards the Detoxome of *Fasciola hepatica* (P67)*

Rebekah Stuart1, Paterson S2, Morphew R1, Brophy P.M1
1 Institute of Biological, Environmental and Rural Sciences, Aberystwyth University, Wales, UK. SY23 3GF. 2Institute of Integrative Biology, University of Liverpool, Biosciences Building, Crown Street, Liverpool, UK L69 7ZB.

Fasciolosis of livestock is a global threat to food security and is now an increasing food borne risk to humans. At present there are currently no commercial vaccines to underpin control programmes. Therefore, reported treatment failures and resistance to Triclabendazole (TCBZ) is of particular concern as TCBZ is the only anthelmintic with activity against both mature and pathogenic immature fluke. To secure future anthelmintic control of fasciolosis, uncovering the parasites full detoxification capacity must be of paramount importance. This will not only provide potential strategies to chemically modify current TCBZ compounds but also provide biomarker panels to measure detoxification pathways correlated to resistant parasites. Therefore, we have mined a draft *Fasciola hepatica* genome and several transcript data sets to reveal the anthelmintic detoxification capacity of the parasite. We report for the first time, expression of the Phase I drug detoxification Cytochrome P450 (CYP450) superfamily in adult *F. hepatica*. We have characterized two novel CYP450s from *F. hepatica* and hope to readdress the perceived over estimation of the importance placed on the role of the Phase II detoxification protein superfamilies.

Limiting Damage during Infection: Lessons from Infection Tolerance for Novel Therapeutics (P68)

Pedro Vale1, Andy Fenton2, Sam P. Brown1
1 Centre for Immunity, Infection, and Evolution and Institute of Evolutionary Biology, School of Biological Sciences, University of Edinburgh, Edinburgh, UK, 2 Institute of Integrative Biology, University of Liverpool, Liverpool, UK

The distinction between pathogen elimination and damage limitation during infection is beginning to change perspectives on infectious disease control, and has recently led to the development of novel therapies that focus on reducing the illness caused by pathogens ("damage limitation") rather than reducing pathogen burdens directly ("pathogen elimination"). While beneficial at the individual host level, the population consequences of these interventions remain unclear. To address this issue, we present a simple conceptual framework for damage limitation during infection that distinguishes between therapies that are either host-centric (pro-tolerance) or pathogen-centric (anti-virulence). We then draw on recent developments from the evolutionary ecology of disease tolerance to highlight some potential epidemiological and evolutionary responses of pathogens to medical interventions that target the symptoms of infection. Just as pathogens are known to evolve in response to antimicrobial and vaccination therapies, we caution that claims of "evolution-proof" anti-virulence interventions may be premature, and further, that in infections where virulence and transmission are linked, reducing illness without reducing pathogen burden could have non-trivial epidemiological and evolutionary consequences that require careful examination.

The genomes and transcriptomes of tapeworms (P69)

Magdalena Zarowiecki1, Jason Tsai1, Nancy Holroyd1, Alejandro Sanchez Flores1,Ferenc Kiss2, Uriel Koziol2, Peter Olson3, Klaus Brehm2, Matt Berriman1

1 Parasite Genomics, Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SA, UK 2 University of Würzburg, Institute of Hygiene and Microbiology, D-97080 Würzburg, Germany 3 Department of Life Sciences, The Natural History Museum, Cromwell Road, London SW7 5BD, UK

We have performed Illumina sequencing of the transcriptomes from 18 representative life-stages of several species of tapeworms, with the aim of understanding how these species develop and function in different environments. In particular, we are looking at how the parasite expression *in vivo* differs from that in the host, if the parasite is using aerobic or anaerobic metabolism in its host, how the stem cells allow the species to proliferate by budding, and if RNA-Seq can explain morphological differences in the genetically very similar species *Echinococcus multilocularis* and *E. granulosus*. The thorough sampling of RNA-Seq is also used to predict alternative splicing patterns, including tapeworms-specific spliced-leader mediated *trans*-splicing. Associated deep sequencing of miRNA is used to narrowing down the list of potential miRNA targets. Pathway information is cross-linked with clustering analysis to predict co-expressed and potentially interacting genes. Together, this will give us a deeper understanding of specific and general patterns of gene-expression in tapeworms, and give us targets for drugs, diagnostics, and for understanding host-parasite interactions.

Effects of opportunistic bacteria on the demography of *Caenorhabditis elegans* (P70)

Eric Mooring, S. Anaid Diaz, Olivier Restif

Department of Veterinary Medicine, University of Cambridge, Madingley Road, Cambridge, CB3 0ES, UK

While *Caenorhabditis elegans* is a well-studied model organism in molecular and developmental biology and has been used as a model system for studying innate immunity and host-pathogen interactions, relatively little is known about its ecology. Because of the potential utility of *C. elegans* as a model system for host-pathogen ecology and evolution research, it is important to understand the effects of different bacterial species on *C. elegans* mortality and fecundity. We conducted a full factorial experiment in which nematodes were raised either singly or in cohorts of 10 or 25 individuals and fed *Escherichia coli* OP50-1, *Pseudomonas aeruginosa* PAO1, or *Salmonella enterica* serovar *Typhimurium* JH3010. Nematodes were transferred to fresh plates daily, and we recorded when nematodes died and when and how many viable eggs were laid. We found that diet had a significant effect on survival, while cohort size did not. Additionally, we calculated asymptotic population growth rates for each combination of treatments. We found that the highest growth rates generally corresponded to nematodes fed *S. enterica*, despite the fact that nematodes fed *S. enterica* experienced premature mortality relative to nematodes fed *E. coli*. The effect is due not only to differences in overall fecundity, but also due to earlier fecundity in nematodes fed *S. enterica*. This result demonstrates the importance of including the timing of fecundity when assessing the effects of pathogens on overall nematode fitness.

The BBSome subunit BBS1 is required for host infectivity in *Leishmania major* (P71)

Helen Price, Daniel Paape, Deborah F. Smith

Centre for Immunology and Infection, University of York, YO10 5DD. UK

Bardet-Biedl syndrome (BBS) is a human genetic disorder with a spectrum of symptoms caused by primary cilium dysfunction. The disease is caused by mutations in one of at least 16 identified genes, of which 7 encode subunits of the BBSome, a protein complex required for specific trafficking events to and from the primary cilium. The molecular mechanisms associated with BBSome function remain largely unknown. We have generated null and complemented mutant lines of the BBSome subunit BBS1 in *Leishmania major*. The BBS1 null parasites have no apparent defects in growth, motility or differentiation *in vitro* but accumulate vacuoles at the flagellar pocket. Infectivity of these parasites for macrophages *in vitro* is reduced compared to wild type controls but the mutant parasites retain the ability to differentiate to the intracellular amastigote stage. However, infectivity of BBS1 null parasites is severely compromised in a BALB/c mouse footpad model. We hypothesise that the absence of BBS1 in *Leishmania* leads to defects in specific trafficking events that affect amastigote persistence in the host. This is the first report of an association between the BBSome complex and pathogen infectivity, providing a tractable model for the study of this complex in both *Leishmania* and human cells.

Characterization of nematode parasite specific acetylcholine receptor as potential target for the development of novel anthelmintics (P72)*

Elise Courtot1,2, Claude L. Charvet1,2, Jacques Cortet1,2, Nicolas Peineau3, Adrian J. Wolstenholme4, Debra J. Woods5 and Cédric Neveu1,2

1 INRA, UMR1282 Infectiologie et Santé Publique, F-37380, Nouzilly, France, 2 Université de François Rabelais de Tours, UMR1282 Infectiologie et Santé Publique, F-37000, Tours, France, 3 Université François Rabelais de Tours, Département de physiologie animale, F-37000, Tours, France, 4 Dept. Of Infectious Disease & Center for Tropical and Emerging Global Disease University of Georgia Athens, GA 30602, USA, 5 Veterinary Medicine Research and Development, Zoetis, 7000, Kalamazoo, MI 49001, USA

Acetylcholine receptors of parasitic nematodes (AChRs) represent major targets of widely used anthelmintics. AChRs are ion channels located at the neuromuscular junction involved in fast synaptic neurotransmission, made of 5 identical subunits (homopentamers) or 5 different subunits (heteropentamers). There is a large repertoire of genes encoding for AChR subunits in nematodes (25 in the model nematode *Caenorhabditis elegans*), and the aim of this present project is to investigate the AChR diversity in parasitic nematode to identify and characterize novel drug targets. Based on genomic data analyses we have identified two AChR subunits that appear to be specific to mammalian parasitic nematodes and named ACR-26 and ACR-27. These two subunits are shared by the clade V trichostrongylid species as well as the clade III ascarid and filarid species. The full-length cDNA of these genes were cloned from the trichostrongylid nematode *Haemonchus contortus*. We report that these two subunits are able to form a functional AChR when expressed into Xenopus oocytes. The detailed pharmacological characterization of this receptor revealed some very attractive properties. These results open the way for further investigations of the AChR diversity in parasitic nematodes and provide a solid basis for the development of novel anthelmintic compounds specific for mammalian parasites.

Relationships between virulence, replication and the host phylogeny following a host shift of an RNA virus (P73)

Ben Longdon, Jon Day, Frank Jiggins

Department of Genetics, University of Cambridge

Emerging viral diseases are often the result of a host shift, where the virus originates from a different host species. Pathogen virulence (the harm a pathogen does to its host) following a host shift can be extremely high, as seen in several high profile cases. However, many host shifts presumably go undetected due to the pathogen causing little or no virulence in its new host. Here, we test whether virulence is predicted by the host phylogeny following an artificial host shift. We have carried out a large cross infection experiment using 50 species of *Drosophilidae*, and a natural RNA virus (DCV) to examine patterns of virulence across the host phylogeny, and how virulence correlates with viral replication.

Molecular Characterisation of Hammondia oocysts from the Arabian red fox (*Vulpes vulpes arabica*) in Saudi Arabia.(P74)

Osama B. Mohammed, Sawsan A. Omer, Abdulaziz N. Alagaili

Department of Zoology, College of Science, King Saud University, P. O. Box 2455, Riyadh 11451, Saudi Arabia.

The Arabian red fox (*Vulpes vulpes arabica*) shed upsorulated oocysts of a Hammondia sp. after consumption of meat from the Mountain gazelle (*Gazella gazella*) contained cysts related to a different cyst forming coccidia. Dogs and cats

received the same meat and did not shed unsporulated oocysts or other stages. The oocysts were spherical to subspherical which sporulated in 3 days and they measured 10.9 ± 1.4 X 10.1 ± 1.3 µm. Each oocyst contained 2 sporocysts which measured 6.0 ± 0.6 X 4.7 ± 0.8 µm each with four sausage shaped sporozoites. The sporulated oocysts shed by the Arabian red fox were different from all isosporan parasites infecting the red fox. However, it was morphologically related to *Isospora triffitae*. It was also morphologically similar to *Hammondia heydorni* and *Neospora caninum* of the dog. Molecular identification using the LSU rDNA and the alpha tubulin genes indicated that the oocysts recovered from the Arabian red fox is distinct from the *Hammondia heydorni* isolates from the dog. The oocysts shed by the Arabian red fox are probably related to *Hammondia triffittae*.

Toxoplasma gondii infections in Libya (P75)*

Muftah Saleh Abushahma, Geoff Hide, Mohamed Benhasin
Libya, UK

Toxoplasma gondii is a parasite discovered in 1908 by Nicolle and Manceaux; they found a protozoan in tissue of a hamster-like rodent in laboratory of the Charles Nicolle at the Pasteur Institute in Tunisia. It is an obligate intracellular protozoan parasite, capable of infecting most species of warm-blooded vertebrates including mammals and birds. This infection is world-wide and one third of the human population is infected chronically. Toxoplasmosis is an important disease and a causative agent that is responsible for abortion in human and farm animals. *T. gondii* can be transmitted in three ways: via cat faeces, eating raw meat and congenital transmission (mother to child). One hundred and fifty mothers were recruited to the study producing 153 children , of these a ratio of male to female of 1.22:1, giving a slightly higher bias than the Libyan average (1.05:1). Mother's ages ranged from 18 – 45 with an average age of 28 years. The sample contained mothers having their first child to those who had 11 children. The pregnancy success of this cohort was 8 deaths per 145 live births (55.1/1000 live births) which is higher than the Libyan national average (21.9/1000) and the UK national average of 4.93/1000. Overall, the samples set did not appear biased when compared with Libyan national statistics. Miscarriage was analysed with respect to various parameters. As has previously been reported, success of pregnancy declined with increasing with age although this was more marked in mothers older than 38.

Analysis of several putative replication restart factors in _Trypanosoma brucei_ DNA repair and VSG switching (P76)*

Rebecca Devlin, Richard McCulloch
Wellcome Trust Centre for Molecular Parasitology, University of Glasgow

The primary mechanism of variant surface glycoprotein (VSG) switching in *Trypanosoma brucei* is gene conversion by homologous recombination. However, the initial stages of VSG switching are unclear. We are investigating the hypothesis that DNA replication stalling occurs upstream of the VSG gene in a region known as the 70 bp repeats and that this precipitates elevated levels of DNA double strand breaks, leading to VSG switching. To test this, we are investigating whether four putative replication restart factors are involved in DNA repair, in particular acting on stalled replication forks, and if they contribute to VSG switching. We describe the activities of two helicases; one RecQ helicase and a Pif-1 family helicase, PIF6. recq2 and pif6 null mutants were viable and displayed differing phenotypes with respect to survival after exogenous DNA damage. Interestingly, both null mutants exhibited decreased survival after the induction of a DNA double-strand break (DSB) internal to the chromosome. In contrast, pif6 null mutants displayed increased survival relative to wild type cells when the break was located adjacent to the transcribed VSG. A third factor, the structure-specific endonuclease MUS81, was also analysed. mus81 null mutants were viable and null mutants displayed increased susceptibility to DNA damaging agents. These data suggest that RECQ2, PIF6 and MUS81 play a role in the repair of DNA damage and we are currently establishing an assay to investigate a possible role in VSG switching.

Genetic diversity in natural _Trypanosoma congolense_ population (P77)*

Eliane Tihon, Imamura H, Dujardin JC, Van Den Abbeele J
Institute of Tropical Medicine Nationalestraat 155 B-2000 Antwerpen Belgium

Isometamidium Chloride (ISM) is one of the principal drugs used to counteract *Trypanosoma congolense* infection in livestock, both as a prophylactic as well as a curative treatment. Numerous cases of ISM resistance have been reported in different African regions, representing a serious problem in the battle against animal African Trypanosomiasis (AAT). To discover genetic signatures associated with ISM resistance in *T. congolense*, the whole genome of a total of 45 ISM-resistant and sensitive strains of various AAT endemic regions have been sequenced using an Illumina MiSeq platform.

Subsequently, the obtained reads were mapped to the reference genome *T. congolense* IL3000. First results identified 23 793 712 SNPs among the Savannah sub-group, of which 817 072 singleton SNPs, indicating a high genetic diversity in natural *T. congolense* population. Preliminary coding SNPs analysis highlights a group of resistant strains with more genes with SNPs under positive selection, which could be associated with drug resistance. No significant changes in genes copy number or ploidy were observed in the different strains. Experimental ISM-inductions in the murine host are currently ongoing and will allow us to better understand the relevance of the genomic analysis results.

The isolation and identification of potential vaccine antigens against poultry red mite (P78)*

James Pritchard1, Fiona Tomley1, Olivier Sparagano2
1The Royal Veterinary College, University of London, Hawkshead Lane, North Mymms, AL9 7TA 2Northumbria University, Faculty of Health and Life Sciences, Newcastle upon Tyne, NE1 8ST

The poultry red mite is the most economically important ectoparasite affecting laying hens throughout the world. Bird welfare suffers due to red mite blood feeding and these mites are also implicated as vectors of diseases such as Newcastle disease, Salmonella and Equine encephalitis. Current acaricidal control is not sufficiently effective due to increased resistance thus alternative control strategies are required. We aim to isolate concealed antigens from the gut of mites and test their efficaciousness as potential vaccine candidates. Effective natural immunity has been previously demonstrated from the tick vaccine TickGARD™ using concealed antigens against the cattle tick *Rhipicephalus (Boophilus) microplus*. Using this concept we focus specifically on the isolation of membrane proteins of the mite gut which are both concealed and are also presented to antibodies when a blood meal occurs once a poultry bird has been immunised. Current protocols are based on separating membrane proteins via low speed differential centrifugation. This has provided distinct protein fractions validated via western blots with already defined anti-Cathepsin D and Histamine Release Factor antibodies. Mass spectrometry and comparison of these samples to our red mite transcriptome library has revealed several red mite proteins of interest. Antibody libraries specific to these isolated proteins will be created by biopanning and should help to identify the location of the expression of our potential vaccine targets via immunohistochemistry. An in-house mite collection and isolation protocol has been established and a protocol for sectioning mites is also showing early promise to be used for our future studies.

Ivermectin resistance in UK field populations of *Haemonchus contortus* (P79)

Roz Laing 1; Axel Martinelli 2; Kirsty Maitland 1; Lenka Lecova 3; Charlotte Burgess 4; Andrew Rezansoff 5; Libby Redman 5; Phil Skuce 4; James Cotton 2; John Gilleard 5; Andrew Tait 1; Eileen Devaney 1
1 MVLS, University of Glasgow, UK; 2 Wellcome Trust Sanger Institute, Cambridge, UK; 3 Charles University of Prague, Czech Republic; 4 Moredun Research Institute, Edinburgh, UK; 5 University of Calgary, Alberta, Canada

Anthelmintic resistance is a major threat to the UK sheep industry and is an emerging concern for parasite control in other species. The mechanisms underlying resistance in parasitic nematodes are not fully understood and current recommendations for sustainable parasite control are based largely on theory. We are investigating genetic changes associated with ivermectin resistance (IVM-R) in the sheep parasite *Haemonchus contortus*, using both candidate gene and whole genome approaches, to improve our understanding of how resistance arises and spreads. SNPs in numerous candidate genes have been associated with IVM-R, but the relevance of these mutations to resistance in the field remains unclear. We examined UK field populations of *H. contortus*, differing in ivermectin treatment history, for evidence of selection at candidate gene loci (glc-5, avr-14 and lgc-37) using capillary sequencing and RFLP, combined with microsatellite marker analysis. High levels of polymorphism were identified at all loci and a degree of population sub-structuring was apparent. These factors can confound candidate gene analysis and may underlie the plethora of genes associated with IVM-R to date. Our second, global approach is to use RAD-seq to genotype individual worms from UK farm populations with differing anthelmintic regimes to identify markers associated with ivermectin selection. We are also genotyping *H. contortus* larvae from IVM-susceptible and IVM-resistant laboratory isolates and two IVM-resistant backcrosses. This approach does not rely on prior assumptions as to the mechanism of IVM-R and provides genome-wide coverage of markers, which will be used to identify regions of the genome under selection.

Cytokine and Chemokine Analysis in Aqueous Humour of Patients with Trematode Induced Anterior Chamber Granulomatous Uveitis (P80)*

Lalan Kumar Arya Non1, SR Rathinam 2, Lalitha Prajna1 ,Usha Kim2 ,Veena Tandon3
1 Department of Microbiology, AMRF, Madurai, 2 Department of Uvea, AEH, Madurai and Department of Orbit, AEH,

Madurai 3 Department of Parasitology, North Eastern Hill University, Shilong, Meghalaya, India

Anterior chamber granulomatous uveitis in children of South India is one of the newly recognized ocular diseases. It is a major infectious cause for paediatric uveitis and is potentially vision threatening condition. The etiological agent of the disease was confirmed as a trematode and the aqueous fluid cellular analysis revealed a predominance of eosinophil in our studies (JAMA Oph 2012). The proposed research aim was to analyse the aqueous humour cytokine and chemokine profile in patients with trematode induced granulomatous uveitis. We included nine new patients with Trematode induced granulomatous uveitis along with 8 patients with various forms of human endogenous uveitis, associated with VKH disease (3), HLA-B27 (2), Fuchs (1) and non granulomatous (2). Patients with no ocular pathology other than cataracts were enrolled as non-inflammatory controls (9). Using a multiplex assay, we determined the concentrations of 17 cytokines and chemokines in aqueous humour specimens obtained from patients with various groups. The trematode induced uveitis showed significantly higher levels of IL-1β, IL-6, IL-8, MIP-1α, MIP-1β, & RANTES followed by IL10 & IL-13 & TNF-α in comparison with other endogenous Uveitis and cataract control groups. The levels of IL-5 in trematode induced uveitis was just double than other Uveitis. Kruskal Wallis analysis revealed significant differences in aqueous cytokine and chemokine concentrations among the various group. Proinflammatory response was highly significance in patients with trematode induced granulomatous uveitis in comparison with endogenous uveitis & cataract controls groups. There was mixed Th1 & Th2 response in trematode induced granulomatous Uveitis.

Micro-volume leucodepletion of malaria-infected blood samples (P81)*

Mihir Kekre, Daniel Mead, Eleanor Drury, Dominic Kwiatkowski, Bronwyn MacInnis, Susana Campino
Wellcome Trust Sanger Institute, Hinxton, Cambridge, United Kingdom CB10 1SA

The mutational plasticity of the *P. falciparum* genome causing drug resistance can be best unravelled using whole genome sequencing (WGS) technology. The primary obstacle to WGS parasite DNA directly from whole blood is the overwhelming contamination of human leucocyte DNA (typically >99% of the sample). Conventional cellulose columns, utilized in laboratories and endemic field sites to remove white blood cells, only work efficiently at sample volumes of above 1ml and at parasitemias of >5,000 parasites/µl (0.1%). Here we introduce two novel blood filtration methodologies designed exclusively for micro-volume (0.05-0.2ml) patient samples. Under simulated field conditions, we tested the efficacy of a new cellulose powder variant and a novel solid cellulose column and captured above 95% and 90% of human DNA from parasite-infused whole blood. This reduction was independent of parasitemia, with sequencable parasite gDNA recovered at parasitemias as low as 1500 parasites/ µl (0.03%). Both methods are inexpensive, easy to assemble, stable for long-term storage and require no operator lab expertise. These methods will permit sequencing parasites from blood volumes that need but a finger prick, making it faster than venous sampling. It will also eliminate struggle for patient blood at multi-study field sites, aid rapid sample processing and be minimally invasive to ailing volunteers, especially children. The features incorporated into the device's current design and working as well as envisioned field-adaptable modifications should make it robust enough for high-throughput sampling and effective global malaria surveillance.

MicroRNA regulation of development in *Haemonchus contortus* (P82)*

Neil Marks1, Alan Winter 1, Brett Roberts 1, Henry Gu 1, Axel Martinelli 2, Eileen Devaney 1, Collette Britton1
1 Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, G61 1QH 2 Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge, CB10 1SA, UK

MicroRNAs (miRNAs) are short, non-coding RNAs that regulate gene expression at the post-transcriptional level. miRNAs play important roles in cell and tissue development and are associated with a number of diseases (e.g. cancer, heart disease). In the free-living nematode *Caenorhabditis elegans*, miRNAs are essential for correct developmental progression. We hypothesise that miRNAs are important regulators of development in the parasitic nematode, *Haemonchus contortus*. 195 miRNAs were discovered in *H. contortus* by deep-sequencing and bioinformatic approaches. Microarray analysis identified 47 miRNAs with significant variation in expression between life-cycle stages (L3 to adult), 16 of which are specific to nematodes. A number of miRNAs are also enriched in adult gut tissue. To identify possible targets and functions of miRNAs of interest, we are using bioinformatic and experimental approaches. Various prediction algorithms identified potential miRNA binding sites in the 3' UTRs of *H. contortus* and *C. elegans* mRNAs. Targets common to both species were studied using qPCR and *C. elegans* mutants. Inverse developmental expression patterns between miRNA and mRNA were suggestive of a possible interaction. We are currently adapting cross-linked

immunoprecipitation (CLIP) to pull-down mRNA-miRNA effector complexes. Together these data will confirm miRNA-mRNA interactions, allowing us to identify developmental pathways under miRNA regulation. Targeting these miRNAs or key target genes may provide a novel therapeutic approach.

Hide and Spread for intracellular *Leishmania* parasites - from infection to apoptosis (P83)*

Rajeev Rai, Lawrence Harbige, Giulia Getti
School of Science University of Greenwich, Medway, Kent ME4 4TB UK

Leishmania is a protozoan parasite responsible for human disease called leishmaniasis. The parasite life cycle alternates between two stages. The promastigotes stage develops inside the sandfly vector and once injected into a mammalian host, enters macrophages and establish itself as intracellular replicating amastigotes. The dissemination of amastigotes to uninfected macrophages is crucial for the development of the disease. However, the molecular mechanism of spreading is largely unknown. Natural infection studies in mice have shown that early phase of infection occurs "silently" without accompanying immunopathological changes. A process resembling cell death via apoptosis, which involves the removal of damaged cells by macrophages without eliciting an inflammatory response, speculated us that *Leishmania* parasites could leave the host cell within apoptotic bodies, evading the host immune defense system. Our study investigates apoptosis in terminally differentiated human THP-1 cells following infection with four GFP expressing species of *Leishmania* parasites. Through flow cytometry, infection and early apoptotic features were detected simultaneously for four days both on apoptotic induced and non-induced cells. Our results revealed a significantly higher level of early apoptosis upon parasite infection compared to uninfected cells. Remarkably, within 72 and 96 hours from infection, apoptotic induction caused up to a 4 fold increase in percentage of infected cells when compared with non-induced cells. Moreover, an increase in the level of apoptosis was also detected, further demonstrating a link between infection and apoptosis. Overall, the results support the hypothesis that apoptosis induction could participate in parasites spreading between human host cells.

Optimisation of the delivery of Foreign genes using *Eimeria* species parasites as novel vaccine delivery vectors (P84)*

Elaine Pegg1, Virginia Marugan-Hernandez1, Sarah Macdonald1, Keith Redhead2, Colin Crouch2, Ian Tarpey2, Michael Francis2, Damer Blake1, Fiona Tomley1.
1 *The Royal Veterinary College, University of London, Hawkshead Lane, North Mymms, AL9 7TA, UK, 2 MSD Animal Health, Walton Manor, Milton Keynes, MK7 7AJ*

The poultry industry is one of the largest agricultural industries in the UK, accounting for up to 40% of the UK meat market. Due to the demanding nature of poultry production commercially reared chickens are also among the most profoundly vaccinated livestock, leading to a drive in the development of new vaccines to combat the spread and subsequent cost of disease within the industry. The vaccination of poultry against *Eimeria* species parasites, which cause coccidiosis, primarily relies on the use of live parasites. The recent development in techniques to transfect *Eimeria* has offered the opportunity to use these live vaccines to deliver additional vaccine antigens against a range of pathogens including *Campylobacter jejuni. Campylobacter jejuni* is a gram-negative, zoonotic bacteria and one of the leading causes of foodborne illness worldwide. The ability of *Eimeria tenella* to successfully express endogenous antigens was previously shown using anti-C. jejuni vaccine candidate CjaA, which induced immune protection against *C. jejuni* compared to unvaccinated and *E. tenella* wild-type controls. One of the aims of the project is to optimise expression by *E. tenella* of anti-*C. jejuni* vaccine candidates. If successful, pre-existing live anticoccidial vaccines based on attenuated species of *Eimeria* have commercial potential to be used as vectors, inducing immunity against *Eimeria* as well as against heterologous antigens derived from other pathogens.

A novel kinesin involved in flagellum attachment and positioning in *Trypanosoma brucei* (P85)

Linda Kohl1, S. Luiggi1, A. Raïa1, L. Petit1, T. Blisnick2, L. Pao1, P. Bastin2, P. Grellier1
1 *Biodiversity and Adaptation of Eukaryotic Microorganisms to their Environment, UMR7245 MNHN/CNRS National Museum of Natural History, Paris, France, 2 Trypanosome Cell Biology Unit, Institut Pasteur & CNRS URA2581, Paris, France*

Kinesins are motor proteins that transport cargo along microtubules using ATP. KIN5 is an orphan kinesin, i.e. it does not belong to any of the known kinesin families, and it is found only in trypanosomatids. The function and localisation of this protein are investigated in *Trypanosoma brucei*, which possesses a single attached flagellum. Using a YFP::KIN5 fusion,

we show that in procyclic cells KIN5 is localised in the flagellum (probably the axoneme), with a strong fluorescent signal at the distal tip. Depletion of KIN5 by inducible RNA interference results in cells with a mis-positioned and partially detached flagellum. The displacement of the flagellum and its associated structures results in a rearrangement of several organelles, such as the nucleus, the kinetoplast and the endocytic compartment. The flagellum of the KIN5RNAi cells is still beating, but the cells are unable to swim. Preliminary data indicate that the filament of the Flagellum Attachment Zone is smaller in length in cells depleted of KIN5. Intriguingly, the KIN5RNAi mutants grow almost normally in culture, indicating that the cells have adapted to the partially detached flagellum, probably by the modifications of the intracellular organisation. We are now investigating the relationship between KIN5 and other proteins known to be involved in flagellum attachment.

Circulating microRNAs are potential diagnostic biomarkers of filarial infections (P86)

Lucienne Tritten1, Andrew Moorhead 2,3, Erica Burkman 2,3, Mohammed Satti 4, James Geary 4, Charles Mackenzie 4 and Timothy Geary 1

1 Institute of Parasitology, Centre for Host-Parasite Interactions, McGill University, 21,111 Lakeshore Road, Sainte-Anne-de-Bellevue, Quebec H9X 3V9, Canada, 2 Department of Infectious Disease, College of Veterinary Medicine, University of Georgia, Athens, Georgia 30602, USA, 3 Filariasis Research Reagent Resource Center, College of Veterinary Medicine, University of Georgia, Athens, Georgia 30602, USA, 4 Department of Pathobiology and Diagnostic Investigation, College of Veterinary Medicine, Michigan State University, East Lansing, MI 48824, USA.

Filarial nematodes cause chronic and profoundly debilitating diseases in both humans and animals. microRNA (miRNA) deep-sequencing combined with bioinformatics revealed >200 unique mature miRNA sequences of nematode origin in *Dirofilaria immitis*-infected dog plasma, and 21 in *Onchocerca volvulus*-infected human serum. In search of new potential biomarkers of infection, we explored the applicability of circulating miRNAs released by filarial nematodes into their hosts' bloodstream for detection of infection by RT-qPCR. Total RNA samples obtained from *D. immitis*-infected dog plasma samples were subjected to stem-loop RT-qPCR assays for amplification of two parasite-derived miRNAs (currently named dim-miR-71 and dim-miR-34). Both assays efficiently discriminated infected from uninfected samples, where no specific miRNA amplification occurred. The assays also distinguished between *Brugia pahangi*-infected and healthy dogs, as the miRNA sequences are identical in both parasites. However, absolute miRNA copy numbers were not well correlated with the corresponding microfilaria counts. Using a highly sensitive detection method, miRNAs represent potential diagnostic biomarkers for filarial parasites, even for those that do not reside in the bloodstream.

An RNAi screen uncovers the major determinants of human serum sensitivity in *Trypanosoma brucei brucei* (P87)

Sam Alsford1, Rachel B. Currier1, José Afonso Guerra-Assunção1, Taane G. Clark1, David Horn2

1 London School of Hygiene & Tropical Medicine, Keppel Street, London WC1E 7HT, 2 Division of Biological Chemistry & Drug Discovery, College of Life Sciences, University of Dundee, Dow Street, Dundee DD1 5EH

African trypanosomes cause lethal diseases but display distinct host ranges. *Trypanosoma brucei brucei* causes nagana in livestock but fails to infect humans, while *T. b. gambiense* and *T. b. rhodesiense* cause sleeping sickness in humans. *T. b. brucei* is sensitive to innate immune complexes found in normal human serum known as trypanolytic factor (TLF) 1 and 2. TLF resistance mechanisms of *T. b. gambiense* and *T. b. rhodesiense* are now known to arise through either gain or loss-of-function, but our understanding of factors that render *T. b. brucei* susceptible to TLF remains incomplete. A genome-scale RNA interference (RNAi) library screen for reduced sensitivity to human serum identified only four high-confidence 'hits', including all three genes previously shown to sensitize *T. b. brucei* to human serum: the haptoglobin-haemoglobin receptor, inhibitor of cysteine peptidase (ICP) and the lysosomal protein, p67. The fourth gene identified encodes a predicted protein with eleven trans-membrane domains. Using chemical and genetic approaches, we show that ICP sensitizes *T. b. brucei* to human serum by modulating the essential cathepsin, CATL, a lysosomal cysteine peptidase. A second cathepsin, CATB, likely to be dispensable for growth in culture, has little or no impact on human-serum sensitivity. Our findings reveal the major determinants of human-serum sensitivity in *T. b. brucei*. They also shed light on the lysosomal protein-protein interactions that render *T. b. brucei* exquisitely sensitive to human serum exposure, and indicate that CATL, an important potential drug target, has the capacity to inactivate TLF.

Smaller, Faster Fluke: muscle-cell calmodulin suppression in juvenile *Fasciola hepatica* (P88)

Paul McVeigh, Erin M McCammick, David J Timson, Angela Mousley, Lance Hammerland, Brenda Bondesen, Nikki J Marks, Aaron G Maule

Institute for Global Food Security, School of Biological Sciences, Queen's University Belfast, Northern Ireland, UK Merial Limited, 3239 Satellite Blvd., Duluth, GA 30096, USA

Fasciola spp. trematodes continue to inhibit the productivity of global ruminant agriculture on a billion dollar scale, and are also responsible for zoonotic infections of an estimated 17 million people in the developing world. The front-line flukicide (triclabendazole) is now blighted by documented incidences of resistance in both veterinary and human infections. Despite these issues, no new broad-spectrum fasciolicides are in development, and no preventative vaccines are available. As a contribution to drug discovery efforts, we have developed RNA interference (RNAi)-based functional assay pipelines for juvenile liver fluke, with which we aim to identify / validate new molecular targets that are essential to liver fluke biology. Amongst our targets of interest are three calmodulin-like genes (CaMs 1-3). These CaMs are expressed in myocytons throughout juvenile worms and have profound RNAi phenotypes that implicate functions in both development and motility. Silencing of CaMs 1-3 yielded juveniles that developed significantly more slowly *in vitro* than untreated/control-treated worms, and also migrated more rapidly through an agar-based motility assay than untreated/control-treated worms. Significantly, both of these RNAi phenotypes were recapitulated following treatment with trifluoroperazine, a compound with antagonistic effects on calmodulin-Ca2+ binding, providing independent validation of the RNAi data. These findings represent the first studies of CaM function in whole fluke and suggest that CaMs represent valid targets for novel fasciolicides. Experiments aiming to establish the impacts of these phenotypes on liver fluke infectivity *in vivo* are ongoing.

Investigations into the (2-aminoethyl)phosphonate Pathway in *Trypanosoma cruz*i (P89)*

Ross Coron

The Terry K. Smith Research Group, Biomedical Science Research Complex - Level 3, The University of St Andrews, North Haugh, St Andrews, Fife, KY16 9ST UK

Potential drug targets against the intracellular protozoan parasite *Trypanosoma cruzi* (causative agent of American Trypanosomiasis) include the biosynthesis of the glycosylphosphatidylinositol (GPI) anchor. The phosphatidylinositol containing glycosylinositolphospholipids (GIPLs) are the major surface constituent of *T. cruzi* cells at all stages of their lifecycle. This structure is invariably decorated with an unusual (2-aminoethyl)phosphonate (AEP or ciliatine) moiety on the 6-hydroxyl of glucosamine in the GPI core and is believed to play a role in host infection and persistence. Additionally, AEP can also substitute for phosphoethanolamine in the linkage between the GPI-anchor and the polypeptide chain of mucins. Although the role of GIPLs in *T. cruzi* have not yet been unambiguously defined, they have shown to act as a contributor to virulence. Additionally, the AEP moiety has been demonstrated to be a virulence factor in a number of bacteria including human pathogen *Bacteroides fragilis*. Entirely absent in the closely related *Leishmania spp., Trypanosoma brucei* and in higher eukaryotic organisms including humans, a greater understanding of the poorly characterized (2-aminoethyl)phosphonate pathway may enable the development of parasite-specific enzyme inhibitors leading to novel chemotherapeutics in the amelioration of American Trypanosomiasis. My research focuses on the expression, purification and biochemical characterization of the AEP biosynthetic/biodegradative enzymes through a range of techniques including enzyme assays, protein crystallization, site-directed mutagenesis studies and fragment based library screening, allowing us to validate the AEP pathway as a drug target in *T. cruzi* both chemically and genetically.

Stress-induced protein thiol oxidation in African trypanosomes (P90)*

Kathrin Diederich1, Thomas Ruppert 2, and R. Luise Krauth-Siegel 1

1 Biochemie-Zentrum der Universität Heidelberg, Germany 2 Zentrum für Molekulare Biologie der Universität Heidelberg, Germany

Post-translational modification of cysteinyl residues represents a common mechanism to protect proteins from irreversible overoxidation and for redox-regulation. Trypanosomes lack classical glutathione reductases, but possess a unique trypanothione/trypanothione reductase-based thiol redox metabolism instead. Nevertheless, the parasites contain trypanothione and glutathione in comparable concentrations. Thus, the question arises whether free glutathione – in addition to acting as precursor molecule for the biosynthesis of trypanothione – is used for thiol redox control. In this work we implemented a redox proteomic approach to identify reversibly oxidized proteins. Bloodstream *Trypanosoma brucei*, grown under standard conditions or exposed to exogenous oxidative stresses, were lysed in the presence of N-ethylmaleimide and treated with human glutaredoxin-1 or DTT. This should result in the specific reduction of glutathione-protein mixed disulfides and of total protein disulfides, respectively. Newly generated free cysteine residues were labeled with N-(6-(biotinamido)hexyl)-3'-(2'-pyridyldithio)-propionamide. After affinity purification and

stable isotope dimethyl labeling, proteins were identified by ESI-MS. We obtained 32 probably S-glutathionylated proteins of which eight candidates showed stress-induced increased oxidation. After DTT treatment, a total of 128 proteins were identified and stress-dependent enhancement of oxidation was observed in 30 cases. Quantitative determination of free glutathione released from proteins and derivatized with 2,3-naphthalendicarboxaldehyde confirmed that protein S-glutathionylation occurs in trypanosomes. The modification is induced by various oxidative stresses and is rapidly reversed after stress removal. Current work focuses on the characterization of parasite-specific proteins involved in the mechanism.

Analysis of fibrillarin as a nucleolar marker in *Trypanosoma cruzi* epimastigotes (P91)

Roberto Hernandez, Ernesto Guerrero, Ana María Cevallos, Santiago Martínez-Calvillo, Imelda López-Villaseñor
Instituto de Investigaciones Biomedicas, UNAM. Apartado Postal 70228. Codigo Postal 04510, Mexico DF. Mexico

Ribosome biogenesis is an essential biological process. In eukaryotic organisms, this process is initiated and almost driven to completion in the nucleolus. Our research group is interested in this phenomenon in the pathogenic parasite *Trypanosoma cruzi*. In a canonical manner, *T. cruzi* cells organize a bipartite nucleolus in proliferative developmental stages of epimastigotes and amastigotes. A bipartite nucleolus denotes an ultra-structural conformation of two nucleolar components: dense fibrillar and granular components. Interestingly, the nucleolus is not assembled in trypomastigotes, which are non-proliferative cellular stages of the parasite. Therefore, the nucleolus can be thought of as being developmentally controlled during the life cycle of this species of trypanosomes. As we are interested in the study of ribosome biogenesis and nucleolar formation in this parasite, we have begun an analysis of the molecules involved in several steps of this process. We study the T*. cruzi* fibrillarin, the methyl-transferase functional subunit of snoRNPs. These molecular complexes methylate the pre-rRNA along its maturation pathway. The *T. cruzi* genome encodes two potential fibrillarin genes (i.e., two haplotype pairs within the CL Brenner hybrid strain). The deduced proteins – 30kDa in mass – differ by 40% in their primary structure. Both molecules show a theoretical fold pattern similar to fibrillarin from archaea. It is not known whether these parasite fibrillarins show differences in their functions. We present data on fibrillarin expression and nucleolar localization in epimastigotes.

Electron microscopic study of the developmental cycle *Eimeria intestinalis* Cheissin, 1948 in rabbit (P92)

Hoda Elfayoumi , Hoda M. Elfayoumi 1, Thabt Sakran 1, Gamal A. Ealshawi 2, H.M. Abdel-Haleema 2, Abo Elhadid 2
1 Parasitology Division, Zoology Department, Faculty of Science, Beni-Swef University, 2 Parasitology Division, Veterinary of Medicine, Beni-Swef University

The endogenous cycle of *Eimeria intestinalis* was studied coccidia-free rabbits by transmission electron microscopy. A total of four asexual generations were observed and two types of merozoites were reported. The first type, multinuclear merozoites with more than one nucleus while the second type, mononuclear merozoites with one central nucleus. Gamonts were developed mainly from the third generation merozoites where the mature gamonts were recorded together with the fourth generation schizonts. The fully formed microgamete had nucleus, mitochondrion and two flagella. The mature macrogametes contained wall forming body I and wall forming body II arranged at the periphery with many reserve food materials such as amylopectin granules and lipids droplets. The experimentally infected rabbits shed unsporulated oocysts on the day eight p.i. These were allowed to sporulated which was completed within 60 h at 25 ± 3°C.

Development of a "Nemabiome" Sequencing Assay for the Relative Quantitation of Parasitic Gastrointestinal Nematodes in Cattle (P93)

John Stuart Gilleard, Russell Avramenko1, Elizabeth Redman1, James Wasmuth1, Roy Lewis2 and John S Gilleard1
1 Faculty of Veterinary Medicine, University of Calgary, Alberta, Canada, 2 Merck Animal Health, Canada

Parasitic nematode infections are estimated to cost the US cattle industry $2 billion/year in treatment costs and lost production. Nematodes often occur as mixed species infections, which vary in pathogenicity and drug sensitivity. Fecal Egg Counts (FEC) are used to assess parasite burdens, however, larvae must be inspected for morphological differences to determine species, which is labour intensive. Detection of resistance involves conducting FECs pre- and post-treatment and determining decreases in egg counts. This procedure is not very sensitive and does not yield information of species present before and after treatment. Molecular markers are faster, more sensitive and more accurate than

traditional methods. The rDNA ITS-2 locus is the favoured region due to its appropriate level of species-specific variation. Previous approaches such as qPCR are semi-quantitative, can only detect pre-defined species, and have repeatability and scalability problems. We developed an approach, akin to 'microbiome' sequencing, in which the rDNA ITS-2 region is simultaneously sequenced from thousands of parasites per sample. The assay provides digital data, allowing accurate assessment of relative species quantities per sample. Multiple samples can be sequenced simultaneously in a single Illumina MiSeq run. We established the protocol from sample preparation to data analysis and are currently verifying the sensitivity, specificity and quantitative accuracy using artificially created larval pools from the major cattle nematodes. We are applying the assay to investigate parasite species prevalence in Canadian cattle. This assay will provide a powerful tool to conduct epidemiological studies, surveillance and improve drug efficacy assessments in cattle parasites.

Genetic diversity of Portuguese *Leishmania infantum* strains by microsatellite analysis (P94)

Sofia Cortes1, Katrin Kuhls2, Mónica Nunes3, Carla Lopes3, Marta Marcos3, Luís Cardoso4, Gabriele Schonian5, Lenea Campino1,6, Isabel Maurício7

1 Centro de Malária e outras Doenças Tropicais (CMDT/ Instituto de Higiene e Medicina Tropical (IHMT)/ Universidade Nova de Lisboa (UNL)/ Lisbon, Portugal; 2 Institut für Mikrobiologie und Hygiene, Charité Universitätsmedizin Berlin, Germany; 3 IHMT/ UNL, Lisbon, Portugal; 4 Departamento de Ciências Veterinárias, Escola de Ciências Agrárias e Veterinárias, Universidade de Trás-os-Montes e Alto-Douro, Vila Real; Parasite Disease Group, Instituto de Biologia Molecular e Celular, Universidade do Porto, Oporto, Portugal; 5 Institut für Mikrobiologie und Hygiene, Charité Universitätsmedizin Berlin, Germany; 6 Departamento de Ciências Biomédicas e Medicina, Universidade do Algarve, Faro, Portugal; 7 Unidade de Parasitologia e Microbiologia Médicas (UPMM)/IHMT/UNL, Lisbon, Portugal

Leishmania infantum is the main etiological agent of zoonotic visceral leishmaniasis in the Mediterranean region, including Portugal. Previously, it has been shown that Portuguese strains, within the prevalent MON-1 zymodeme, present substantial genetic diversity. This study used a multilocus microsatellite analysis approach to analyse more comprehensively the genetic diversity within *L. infantum* parasites from different regions of Portugal to investigate its population structure and any association with different clinical forms of the disease, hosts and geographical origin. A set of 14 polymorphic microsatellite markers was used to characterize 136 Portuguese *Leishmania* strains, and 26 strains from other countries (Brazil, Ethiopia, France, Greece, Spain, and Sudan) for comparison. A total of 108 different genotypes were found, which is a level of genetic diversity comparable to other regions. Paraphyly or polyphyly of non-MON-1 zymodemes was detected for MON-24, MON-29 and MON-98 strains analysed, showing the inadequacy of MLEE for epidemiological surveillance. Thirteen non-unique genotypes were suggestive of clonal transmission. However, high inbreeding coefficients were found in STRUCTURE MON-1 sub-populations, and strains with mixed ancestry were identified, suggesting that recombination also plays a role in the epidemiology of *L. infantum* in Portugal. Some but limited geographical differentiation was observed, with groups of strains from the same regions clustering together, particularly those from canine origin for which there was evidence of at least two local independent transmission cycles. Our results present a new perspective of the epidemiology of *L. infantum* in Portugal.

Protein synthesis is required for establishing signal memory during irreversible switching from bloodstream to insect-stage forms of *Trypanosoma brucei* (P95)

Maria Rosa Domingo-Sananes1, Michael Urbaniak2, Michael Ferguson3, Keith R. Matthews1.

1 Centre for Immunity, Infection and Evolution. University of Edinburgh. UK, 2 Division of Biomedical and Life Sciences. University of Lancaster. UK, 3 College of Life Sciences. University of Dundee. UK.

The life cycle of *Trypanosoma brucei* involves several cell differentiation transitions that allow the parasite's transmission, survival and proliferation. One of these transitions, the differentiation of growth-arrested stumpy forms in the mammalian blood into proliferating insect-stage procyclic forms, can be induced synchronously *in vitro* by addition of *cis*-aconitate. Using single-cell analysis by flow-cytometry to follow the expression of the procyclic-stage marker procyclin and cellular DNA content, we confirm that cells commit to differentiation after 1-3 hours of exposure to CA and demonstrate that this transition is an irreversible bistable switch. This irreversibility implies the existence of positive feedback mechanisms that allow commitment to differentiation: the establishment of "memory" of exposure to the differentiation signal. Such mechanisms probably depend on post-translational modifications (e.g. phosphorylation) and/or the synthesis of regulatory proteins. To investigate these possibilities, we tested whether cells can still commit to differentiation in the presence of the reversible protein synthesis inhibitor cycloheximide. We find that protein synthesis is required for establishment of signal memory and normal commitment to differentiation. To characterize changes at

the proteome level during commitment, we performed SILAC phosphoproteomics during the earliest stages of differentiation. We find that several proteins significantly change in abundance and phosphorylation as the cells lock into the differentiation programme, and are thus potentially implicated in commitment to differentiation or in the differentiation process itself.

Evaluating diagnostic tests for helminth infections (P96)

Astrid Erber1,2, Piero Olliaro3, Kevin Marsh1, Trudie Lang1,2
1 Nuffield Department of Medicine, Centre for Tropical Medicine, University of Oxford, UK 2 The Global Health Network, Centre for Tropical Medicine, University of Oxford, UK 3 World Health Organization Special Programme for Research and Training in Tropical Diseases (WHO-TDR), Geneva, Switzerland

Helminth infections like schistosomiasis, onchocerciasis, lymphatic filariasis, and the soil-transmitted helminths place a considerable burden on the world's most disadvantaged populations. Large control programmes, often initiated and driven by the World Health Organization (WHO) and operating within public private partnerships (PPPs), are dedicated to their control and elimination. They increasingly rely on accurate and affordable diagnostic tests. The original helminthiases diagnostics are rather basic direct parasitological methods, that is, microscopic detection and quantification of parasites in patient samples. Improved diagnostics based on molecular methods have been developed comparably recently, and many are in development. Still, microscopic methods are used as reference standards when evaluating new ones, despite their lack of accuracy. Within this context of the need for improved diagnostics, the presented project is looking at the process of development and evaluation of new diagnostics for tropical diseases in the absence of a gold standard, specifically tailored to helminthic diseases. It is designed as an evidence-based medicine (EBM) and mixed-methods approach. Studies within a trial (SWATs) will be performed within ongoing diagnostics evaluation studies for process analysis, and to identify best practices. Subsequently, the process of diagnostics evaluation studies will be mapped out, taking into consideration the requirements at different stages of disease control programmes. The work aims to identify study designs specifically tailored to the context, appropriate statistical frameworks, and suggested endpoints of studies that could be helpful for future research, and in the synthesis of a body of knowledge.

Genetic diversity of African isolates of *Toxoplasma gondii* (P97)*

Mohammed H Alruhaili, Judith E Smith
School of Environment and Life Sciences, University of Salford, Manchester, UK

Toxoplasma gondii is intracellular protozoa parasite and has the ability to infect all warm-blooded animals including humans. While the three clonal lineages predominate in North America and Europe, strains from other regions in the world appear to have more diverse genotypes. Analysis of isolates from South America, Asia and Africa via PCR-RFLP or microsatellite markers reveal that the majority of isolates have type I, II or III alleles, identical to those in the main three lineages. The main aim of this study is to focus on African isolates and investigate their genetic relationship to global strains and the level of variation across multiple loci relative to reference type II and III strains. The study conducted multi-locus nested PCR analysis of *Toxoplasma gondii* samples collected from Africa, which was applied by using eleven different genetic markers distributed across eight chromosomes and the apicoplast genome "SAG1, 5'-SAG2, 3'-SAG2, Alt.SAG2, GRA6, L358, BTUB, SAG3, C22-8, C29-2, PK1 and Apico" to increase the resolution and discriminative power in detecting the genetic diversity between isolates. The analysis across multiple loci revealed a high level of sequence homology between the African isolates and the reference strains that originate form North America. However there were some limited genetic variations among these isolates. It is noted that the growth characteristics of the parasites differ despite this limited genetic diversity.

Tryparedoxin Peroxidases Protect Bloodstream *Trypanosoma brucei* from Iron-Mediated Lysosomal Damage (P98)*

Corinna Hiller1, Diego Benítez 2, Marcelo Comini 2, and Luise Krauth-Siegel 1
1 Biochemistry Center of Heidelberg University (BZH), Heidelberg, Germany 2 Institut Pasteur de Montevideo, Montevideo, Uruguay

In African trypanosomes, detoxification of lipid hydroperoxides is achieved by non-selenium glutathione peroxidase-type enzymes (Px) which obtain their reducing equivalents from the unique trypanothione/tryparedoxin system. Our previous knockout studies revealed that the two cytosolic Px I and II are essential while the mitochondrial Px III is not.

Bloodstream *T. brucei* lacking Px I and II are fully viable in the presence of the antioxidant Trolox but show severe lipid peroxidation and cell lysis within a few hours when the vitamin E analog is removed from the culture medium. Live cell imaging of the mutant parasites fed with fluorescent dextran now revealed that the cells undergo lysosomal enlargement and/or complete staining before they finally lyse suggesting a direct linkage between the cytosolic peroxidases and the organelle. The removal of fetal calf serum from the medium prolonged the short-term lifespan of the mutant parasites after Trolox withdrawal, pointing to serum components as origin of the lethal phenotype. Indeed, the cytosolic peroxidases protect from oxidative damage due to the endocytosis of iron containing proteins. Our data also showed that the lysosome – not the mitochondrion – is the primary source of intracellular oxidative stress. The minute role of the mitochondrial Px III observed for bloodstream *T. brucei* in culture was corroborated by *in vivo* studies. Px III-deficient parasites were as infective as wild-type cells in the mouse model.

Factors influencing success or failure of community case management of malaria with rapid diagnostic tests: a systematic review (P99)

Esmée Ruizendaal, S. Dierickx, K. Grietens Peeters, H. Schallig, F. Pagnoni, P. Mens
Royal Tropical Institute, KIT Biomedical Research Amsterdam, The Netherlands Instituut Tropische Geneeskunde (ITG), Antwerp, Belgium Global Malaria Programme, Geneva 27, Switzerland

Community case management of malaria (CCMm) by community health workers (CHWs) has been developed to increase access to correct treatment. Currently the World Health Organization (WHO) recommends to treat only confirmed malaria cases, rather than presumptive treatment. Although the general consensus is that CCMm with rapid diagnostic tests (RDTs) is a good strategy, no in-depth review of factors enabling or hampering success has been performed. Here we report an extensive review of the literature. Factors studied were test performance by CHWs, execution and interpretation of tests, adherence to test results, effect on morbidity and mortality, community uptake, referral completion, stock-outs, CHW incentives and motivation and cost-effectiveness. 27 articles were included. The effect of CCMm on morbidity and mortality could not be estimated based on studies performed so far. However, CHWs were able to correctly perform RDTs and showed high adherence to test results. Uptake and acceptance by the community was high, although negative tested patients did not always follow-up referral advice. A high level of stock outs and limited information on CHW motivation are main bottlenecks for sustainable implementation. RDT based CCMm was found to be cost-effective for the correct treatment of malaria cases in areas with low to medium malaria prevalence. We conclude that CHWs can deliver high quality care for malaria using RDTs. However, the effect on morbidity and mortality needs to be further investigated. To ensure a long-term successful program, more information is needed about socio-cultural aspects, CHW motivation and logistical aspects like bottlenecks for stock supply.

The P-glycoprotein inhibitor ketoconazole causes a reversion to ivermectin sensitivity in cyathostomins (Nematoda: Cyathostominae) *in vitro* (P100)*

Laura Peachey1, Jacqui B. Matthews2, Gina L. Pinchbeck1, Faith A. Burden3, Nicola A. Stradling3, Jane E. Hodgkinson1
1 Institute of Infection and Global Health, University of Liverpool, Liverpool, UK, 2 Moredun Research Institute, Pentlands Science Park, Edinburgh, UK, 3 The Donkey Sanctuary, Sidmouth, UK

Anthelmintic resistance is a major veterinary and public health issue globally; of most concern is the level of resistance to the macrocyclic lactones. Recent studies have identified a role in resistance for the ATP binding cassette (ABC) drug transporters, P-glycoproteins (P-gps). This study demonstrates the effect of the P-gp inhibitor ketoconazole on the efficacy of ivermectin (IVM) against equid cyathostomin larvae using the larval migration inhibition test (LMIT). Third stage cyathostomin larvae (L3) were cultured from two populations; 1) with recent history of IVM resistance *in vivo* and 2) naive to anthelmintic exposure. The sensitivity to IVM in each group (n=8) was characterised using the LMIT. The IVM LMIT was repeated for each sample with and without the addition of 10µM ketoconazole. Probit analysis was performed on grouped data from each population to give LC-50 values. The LC-50 value for IVM in Populations 1 and 2 was 4.9 and 2.4µg/ml respectively indicating that Population 1 has a resistant phenotype in comparison to Population 2. Addition of 10µM ketoconazole to IVM in Population 1 caused a drop in LC-50 value from 5.8 to 1.6µg/ml. In Population 2 the effect of the addition of ketoconazole was negligible (1.1 to 0.9µg/ml). This study demonstrates that the P-gp inhibitor ketoconazole causes reversion to a sensitive phenotype in IVM-resistant cyathostomins, inferring that P-gps play a role in their resistance to IVM. This work will be corroborated by investigation into P-gp genes and their expression in cyathostomins.

Role of VPAC receptors in the chemotactic responses induced by VIP in promastigotes of *Leishmania (Viannia) braziliensis in vitro* (P101)

Alicia Ponte-Sucre, M. Giammarresi, E. Díaz, W. Alcazar, M. Padrón
Laboratory of Molecular Physiology. IME-UCV, Central University of Venezuela

Leishmania interacts with its surroundings both in the vector and mammalian host. The resulting physiological response, chemotaxis, results in parasite migration for or against the stimuli. The Vasoactive Intestinal Peptide (VIP) is released in the skin by sensory neurons when mechanical or immunological stimuli exist, signaling sensory mechanisms. This occurs during parasite-host interaction, an essential step for the onset of *Leishmania* infection. Our aim has been to study the role of VPAC receptors in the chemotactic response induced by VIP in *L. (V.) braziliensis*. We analyzed the effect of a non-selective antagonist (VIP6-28) in *L. braziliensis* promastigotes. Chemotaxis was explored by the capillary-two chamber technique previously standardized. VIP (10-10 M) reduces significantly cell migration. The chemo-repellent response was blocked by VIP6-28 (10-8 M), suggesting that the effect could be mediated by VPAC receptors. To gain more insight into the mechanism involved we evaluated the effect of VIP (10-10 M) on the parasite plasma membrane potential (Vm) and the changes induced by VIP6-28 (10-8 M). No significant changes in Vm were found. Our results suggest that *in vitro* the chemotactic response of *L. braziliensis* to VIP may be mediated by transmembrane receptors. We speculate that as has been previously described these VPAC receptors, once activated might induce peptide endocytosis to stimulate intracellular mechanisms resulting in the chemotactic response.

Probing the ABC's of drug resistance in *Fasciola hepatica* (P102)

Erin McCammick1, Paul McVeigh1, Angela Mousley1, Nikki J. Marks1, Jane Hodgkinson2, Steve Paterson2 and Aaron G. Maule1
1 Molecular Biosciences:Parasitology, Institute for Global Food Security, School of Biological Sciences, Queen's University Belfast, UK, 2 Institute of Integrative Biology, University of Liverpool, UK.

Fasicola sp have significant impact on human and veterinary medicine. An estimated 17 million people are infected and fascioliasis is designated a neglected tropical disease by the World Health Organisation. Impacts on the health and welfare of livestock are estimated to cost the agri-food industry billions of US$ per annum. The treatment of fascioliasis/fasciolosis relies on triclabendazole (TCBZ), the only flukicide active against both mature and immature stages of *Fasciola hepatica*. Reports of drug resistance highlight the requirement for a better understanding of resistance mechanisms and novel methods for control. This study aims to assess the involvement of multidrug resistance proteins (MDR/MRP) from the ABC transporter superfamily in the susceptibility of *F. hepatica* to TCBZ and its metabolites. Previous work in *Schistosoma mansoni* implicates elevated transcription of two MDR proteins (SMDR2 and SmMRP1) in reduced susceptibility to praziquantel. Here we report 1) an updated bioinformatic identification of at least 22 putative ABC transporters in the *F. hepatica* draft genome, 2) the presence of orthologues of MDR ABC proteins, 3) a concentration-dependent up regulation of FhMRP1 in newly excysted juvenile *F. hepatica* exposed to TCBZ and its therapeutically active sulfoxide metabolite (TCBZ.SO), and 4) the successful triggering of transcript knockdown of both FhMRP1 and FhMDR2 using RNA interference (RNAi) methodology and subsequent effects on survival following drug exposure.

Ribose 5-phosphate isomerase B knockdown compromises *Trypanosoma brucei* bloodstream form infectivity (P103)*

Inês Loureiro1, Joana Faria1, Christine Clayton2, Sandra Macedo Ribeiro3, Nilanjan Roy4, Joana Tavares1, Anabela Cordeiro-da-Siva1,5
1 Parasite Disease Group, Instituto de Biologia Molecular e Celular da Universidade do Porto, Rua do Campo Alegre, 823, 4150-180 Porto, Portugal 2 Zentrum für Molekulare Biologie der Universität Heidelberg, DKFZ-ZMBH cv Alliance, Im Neuenheimer Feld 282, D-69120 Heidelberg, Germany 3 Protein Crystallography Group, Instituto de Biologia Molecular e Celular da Universidade do Porto, Rua do Campo Alegre, 823, 4150-180 Porto, Portugal 4 Ashok & Rita Patel Institute of Integrated Study & Research in Biotechnology & Allied Sciences, New Vallabh Vidyanagar, Dist-Anand, Gujarat-388121, India, 5 Departamento de Ciências Biológicas, Faculdade de Farmácia da Universidade do Porto, Rua de Jorge Viterbo Ferreira, 228, 4050-313 Porto, Portugal

Ribose 5-phosphate isomerase (Rpi) is involved in the non-oxidative branch of the pentose phosphate pathway and catalyses the inter-conversion of ribose 5-phosphate (R5P) and ribulose 5-phosphate (Ru5P). There are two non homologous forms, the A and the B. The presence of type B in trypanosomatids, and its absence in humans, points RpiB as a potential drug target. This study presents a functional characterization of *T. brucei* ribose 5-phosphate isomerase B

(TbRpiB). *In vitro* biochemical studies confirmed TbRpiB isomerase activity, as it can use both R5P and Ru5P as substrates. TbRpiB knockdown by RNAi affected *in vitro* growth of bloodstream forms, but more importantly *in vivo* parasites infectivity since mice infected with induced RNAi clones exhibited lower parasitemia and a prolonged survival in comparison to mice infected with control parasites. Furthermore, *in vitro* and *in vivo* phenotype was reverted when an ectopic copy of *Trypanosoma cruzi* ribose 5-phospahate isomerase was introduced. These results suggest TbRpiB as a promising drug target for African sleeping sickness, since interfering with this protein represents a way to control *in vivo* parasite growth and infectivity.

LEXSY - an efficient work horse in parasitology (P104)

Reinhard Breitling, Mathias Gruen
Jena Bioscience GmbH Loebstedter Str. 80 D-07749 Jena Germany

We have turned a non-pathogenic strain of *Leishmania tarentolae* - the parasite of lizards - into the expression platform LEXSY. LEXSY comes in two principal architectures permitting either constitutive or inducible expression of recombinant proteins with yields of up to 500 mg per litre of culture. Coexpression of up to four proteins simultaneously is possible allowing production of heteromeric complexes or interacting proteins. In addition, an *in vitro* LEXSY, based on L. tarentolae cell extracts, is available for rapid cell-free protein expression. Here we present selected applications of LEXSY including expression of correctly folded and post-translationally processed proteins of various protozoa (*Kinetoplastids, Toxoplasma*) in LEXSY by making use of the close relationship of their cellular machineries; use of LEXSY vectors for manipulation/studies of pathogenic *Leishmania* species such as *L. amazonensis, L. donovani, L. infantum, L. major,* and *L. mexicana*-overexpression of kinetoplastid derived antigens in LEXSY for generation of novel vaccines - cell-free expression and coexpression of kinetoplastid proteins *in vitro* for biochemical and interaction studies - structural biology (X-ray crystallography and NMR) on LEXSY produced proteins.

Protein arginine methylation by LmjPRMT7 is developmentally regulated in *Leishmania major* (P105)

Tiago Rodrigues Ferreira1, Eliza Ferreira1, Tania Defina1, Barbara Papadopoulou2, Angela Cruz1
1 School of Medicine of Ribeirão Preto, University of São Paulo, Ribeirão Preto, Brazil, 2 Research Center in Infectious Diseases, CHUL, Laval University, Quebec, Canada

Protein arginine methylation is a widely conserved post-translational modification performed by arginine methyltransferases (PRMTs). However, its functional role in parasitic protozoa is still under-explored. The *Leishmania major* genome encodes five PRMT homologs, including PRMT7, which is only found in a restricted group of eukaryotes. We have found that both LmjPRMT7 expression and arginine monomethylation of cellular proteins are tightly regulated during promastigote development, reaching minimal levels at the stationary growth phase. On the other hand, procyclic promastigotes and amastigotes displayed elevated LmjPRMT7 expression. Considering this difference, we aimed to evaluate the infection profiles of engineered transfectants. As a remarkably opposite phenotype, the knockout of LmjPRMT7 led to an increased infectivity in mice both *in vitro* and *in vivo*, while LmjPRMT7-overexpressing parasites displayed attenuated virulence. Several RNA-binding proteins (RBPs) were co-immunoprecipitated with LmjPRMT7, and may be considered putative substrates. This indicates a role for arginine methylation in the post-transcriptional regulation of *Leishmania* gene expression. Considering RBPs are well-known mammalian PRMT substrates, our data suggest that arginine methylation may also modulate the interaction between RBPs and their target RNAs in *Leishmania*. We have performed mRNA-seq experiments to compare the transcriptome of LmjPRMT7 transfectants and the quantitative analysis of the differentially expressed genes is underway. We will also perform RNA immunoprecipitation to evaluate the RNA affinity of specific candidate RBPs in the presence or absence of LmjPRMT7, methylated or not. This work is the first study to describe a possible role of *Leishmania* protein arginine methylation in the regulation of gene expression.

DNA repair: How do trypanosomes deal with dead-end protein-DNA complexes? (P106)*

Roberta Carloni1, Achim Schnaufer2, Heidrun Interthal1
1 Institute of Cell Biology, Darwin Building,The Kings Buildings, The University of Edinburgh, Mayfield road ,Edinburgh EH9 3JR, UK, 2 Institute of Immunology & Infection Research, University of Edinburgh , King's Buildings, Ashworth Laboratories, West Mains Road, Edinburgh EH9 3JT, UK

The goal of our research is to understand how *Trypanosoma brucei* repairs the potentially lethal form of DNA damage

that occurs when a topoisomerase cleaves DNA and becomes covalently trapped at the end of a broken DNA strand. Topoisomerase I stalling can be induced by endogenous DNA damage and by anti-cancer drugs such as camptothecin (CPT). Eukaryotic tyrosyl-DNA phosphodiesterase 1 (TDP1) is a DNA repair enzyme that removes covalently trapped topoisomerase I and other 3' adducts from the 3' end of the DNA at DNA strand breaks. *Trypanosoma brucei brucei* TDP1-/- cells are hypersensitive to CPT and accumulate in late S/G2 upon treatment with the drug. The catalytic activity of TDP1 is required for complementation of this CPT sensitivity since expression of a catalytically inactive mutant form of TDP1 not only fails to complement, but further sensitises TDP1-/- cells to CPT. Surprisingly, expressing a TDP1 variant that carries a mutation analogous to the one that causes SCAN1, a human neurodegenerative disease, does not sensitise TDP1-/- cells further to CPT. By using a set of mutant TDP1 proteins in a TDP1-/- background we hope to answer questions concerning TDP1 function that have so far been elusive. TDP1 is the only detectable enzymatic activity present in *T. brucei brucei* bloodstream forms that has 3' tyrosyl DNA phosphodiesterase activity. Interestingly, downregulation of the DNA double-strand break repair enzyme MRE11 increases the CPT sensitivity of TDP1-/- cells suggesting that the two enzymes function in parallel pathways for the repair

Involvement of DNA Mismatch Repair proteins in the oxidative stress response in Trypanosomatids (P107)

Viviane Grazielle Da Silva1, Priscila Carneiro Campos1, Ceres Luciana Alves1, Carlos Renato Machado1, Richard McCulloch2, Santuza Maria Ribeiro Teixeira1

1 Departamento de Bioquimica e Imunologia, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil, 2 The Wellcome Trust Centre for Molecular Parasitology, University of Glasgow, Glasgow, UK

DNA Repair mechanisms are crucial for genetic maintenance and to generate genetic variability. MSH2 is a protein of the Mismatch Repair Pathway (MMR), known to be responsible for the recognition of mismatches that occur during DNA replication. Previous studies have shown the existence of three different MSH2 isoforms in the *Trypanosoma cruzi* population, named TcMSH2A, B and C. Functional characterization of these isoforms provided evidences indicating that they respond differently to oxidative stress after treatment of parasites with genotoxic agents. In order to investigate the function of TcMSH2 and of other components of MMR, we generated Tcmsh2 mutants. Tcmsh2 +/- are more susceptible to H_2O_2 treatment and accumulate more 8-oxo-G in the mitochondrial DNA (kDNA) when compared with WT parasites. Surprisingly, when both msh2 alleles were deleted, *T. cruzi* epimastigotes become more resistant to H_2O_2 treatment. The msh2 null mutants are also more resistant to the highly oxidative environment inside infected macrophages. Similar to the *T. cruzi* insect form, an adaptation to the loss of MSH2 was also observed in the insect form of a *T. brucei* msh2 null mutant. Whereas bloodstream forms of Tbmsh2 knockout are more susceptible to H_2O_2, procyclic forms are more resistant compared to WT. To better understand the involvement of MSH2 with the oxidative stress response in these parasites, we tried to identify MSH2 interacting proteins. Although we were able to confirm that MSH3 and MSH6 are part of heterodimer complexes formed with MSH2, no differences were observed before or after treatment with genotoxic agents.

MHC Class II DQA1 Diversity and Nematode Resistance in Scottish Blackface (P108)*

Nur Mahiza Md Isa , Michael Stear

Institute of Biodiversity, Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences, Garscube Campus, Bearsden Road, Glasgow G61 1QH, UK

Several groups have reported the existence of alleles within the MHC that are associated with enhanced resistance to nematode infection. The aim of this project was to characterise MHC Class II DQA1 diversity and its association with faecal egg counts (FEC). We studied Scottish Blackface sheep from a commercial upland farm in Southwest Strathclyde. All lambs grazed the same field after weaning until the end of the grazing season. There were five consecutive cohorts of 200 lambs and FEC were estimated every 28 days from May until October. DNA was extracted from the buffy coat. Locus-specific primers were used to amplify the second exon of MHC Class II DRB1, DQA1, DQB1, DQA2 and DQB2 loci. PCR products were sequenced on a capillary sequencer. We identified eight DQA1 alleles including a null allele in the Scottish Blackface sheep, with seven previously published alleles and one new allele. The most common allele was the null allele (50%). DQA1 shows lower genetic diversity than the other class II loci. Linkage disequilibrium was strong in the class II region. There were over a million possible haplotypes but only 23 were observed in the more than 900 lambs tested. There was a significant association between the haplotype carrying the CCC55881 allele and the FEC. We conclude that the MHC Class II DQA1 locus was associated with reduced egg counts. Further work is necessary to determine whether DQA1 is the causative locus or a marker for another locus.

The molecular basis of parasitism in the nematode *Strongyloides ratti* (P109)

Vicky Hunt1, Hayley Bennett3, Bernardo Foth3, Nancy Holroyd3, Taisei Kikuchi5, Anna Protasio3, Nadine Randle2, Jason Tsai4, Jonathan Wastling2, Matt Berriman3, Mark Viney1

1 School of Biological Sciences, University of Bristol, Bristol BS8 1UG, UK 2 Institute of Infection and Global Health, University of Liverpool, Liverpool, L3 5RF, UK 3 Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge, CB10 1SA, UK 4 Institute of Molecular Biology, Academia Sinica, Taiwan, R.O.C. 5 Division of Parasitology, Department of Infectious Diseases, Faculty of Medicine, University of Miyazaki, Miyazaki 889-1692, Japan

The *Strongyloides* lifecycle includes a parasitic female-only stage, which inhabits the small intestine of its host, and a facultative, dioecious free-living adult generation. These adult life-cycle stages are genetically identical, so that comparing parasitic and free-living stages offers an almost unique opportunity to discover the molecular adaptations required to be a successful parasitic nematode. We have used quantitative mass spectrometry and RNAseq analyses to compare the proteome and transcriptome of parasitic and free-living females of *S. ratti*. We find that 15% of genes are differentially expressed between these two life stages. Many of the genes with upregulated parasitic expression are physically clustered in the genome. These comprise 2-18 adjacent genes, mostly with likely similar functions. Approximately 20% of the genes in these clusters code for astacins of the zinc metalloproteases family. The largest clusters are mainly CAP domain-containing genes. These gene families are therefore likely to be key to parasitism in *Strongyloides* and possibly other parasitic nematodes.

The development of RNA interference (RNAi) in the parasitic nematode *Teladorsagia circumcincta* as a method for screening vaccine candidates (P110)*

Thomas Tzelos, Jacqui Matthews, Bruce Whitelaw, Dave Knox

Moredun Research Institute

Teladorsagia circumcincta, a major cause of ovine parasitic gastroenteritis in temperate climatic regions, has developed resistance to the major anthelmintic drug classes and this challenges the future control of the parasite. Vaccination is a potential alternative control method since sheep are able to develop protective immunity. Although potential vaccine candidates have been revealed, the increasing gene datasets suggest that vaccine-target selection may be aided by screening methods such as RNAi. This is a reverse genetic mechanism that causes highly specific gene silencing and was initially described in *Caenorhabditis elegans*. The targets selected for knock-down are vaccine candidates for *T. circumcincta* and included: two members of the Activation-associated Secreted Proteins (ASPs); a Macrophage migration Inhibitory Factor-like (Tci-mif-1) and a Surface Associated Antigen gene (Tci-saa-1). The results have shown a successful knock-down only for the ASP targets after 1 hour of soaking in gene-specific double stranded RNA (dsRNA). This illustrates the inconsistency and the target specificity of RNAi in *T. circumcincta* which has been observed in the past with other parasitic nematodes. Inconsistencies were also observed within the ASP targets with the silencing effect not being reproducible after four successful subsequent experiments. A number of parameters that might affect variability were examined and it was found that the storage period of the larvae plays an important role in the consistency of the RNAi results, with the larvae stored for a short period of time being susceptible and the one stored for a long period of time not being susceptible to RNAi.

The current status efficacy of Artesunate/Sulfadoxine Pyrimethamine tablets for the treatment of uncomplicated *Plasmodium falciparum* Malaria in Great Wad Medani Locality, Gezira State, Sudan (P111)

Maha Mirghani Abdalla Maatoug1, Mirghani Abdelrahman Yousif1 , and Bakri Yousif Mohmmed Nour2,3

1 Faculty of Pharmacy, University of Gezira, Was Medani, Sudan, 2 Blue Nile National Institute for Communicable Diseases, University of Gezira, Was Medani, Sudan, 3 Faculty of Medical Laboratory Sciences, University of Gezira, Was Medani, Sudan

IIn Sudan the National Malaria Control Program adopted the use of artesunate + sulphadoxine-pyrimethamine (AS/SP) as the first line of treatment for uncomplicated malaria since 2004. This study was done in Medani town, Gezira State, Central Sudan to evaluate the current efficacy of AS/SP among infected patients within the national monitoring of antimalarial drugs. From October to December 2011, 81 Patients with uncomplicated *P. falciparum* malaria who met the study inclusion criteria were enrolled, treated with AS/SP and monitored for 28 days. The follow-up consisted of a fixed schedule of check-up visits and corresponding clinical and laboratory examinations. On the basis of the results of these assessments, the patients were classified as having therapeutic failure (early or late) or an adequate response to AS/SP

according to the WHO(2005) susceptibility to antimalarial drugs protocol. Blood samples from each patient were taken on Whatman filter paper (3M) on days 0, 7, 14,21 and 28 and also the day when the parasite and symptoms reappeared for Polymerase chain reaction (PCR) to distinguish between a true recrudescence due to treatment failure and reinfection. At the end of the follow up period (28 days) and before the PCR correction, 76/80(93.8%) of the 81 patients enrolled in the study were classified as had adequate clinical and parasitological response (ACPR), two (2.5%) as had late clinical failure (LCF) and late parasitological failure (LPF), two (2.5%) as lost to follow-up and only one patient (1.2%) as had early treatment failure. The two patients whom were considered to have had late clinical failure (LCF) and late parasitological failure (LPF) when subjected to PCR correction. The result revealed that the late clinical failure and late parasitological failure were due to reinfection rather than due to recrudescence. Therefore the parasitological and clinical efficacy of AS/SP was found to be 98.7% (77/79) This study concluded that the first line treatment (AS/SP) for uncomplicated *P. falciparum* is still effective and monitoring its efficacy is recommended annually in all Sudan states .

Polymorphism and selection acting on toll-like receptor 6 in bank vole *Clethrionomys glareous* (P112)

Agnieszka Kloch
Faculty of Biology, University of Warsaw ul. Zwirki i Wigury 101 02-089 Warszawa Poland

Until recently, the model system for studying the genetic aspects of host-parasite coevolution were genes of the major histocompatibility complex (MHC). However, these genes constitute only a part of the acquired immune response and little is known about the evolutionary processes acting on other components of the complex mammalian immune system. Here, I characterize the polymorphism and selection acting on toll-like receptor 6 from bank voles (*Clethrionomys glareolus*). TLR6 is a part of the innate immunity and it recognizes lipopeptides characteristic for Gram positive bacteria (TLR6). In ~800bp sequenced fragment I found 9 SNPs constituting 6 alleles. Six haplotypes were identified, and half of them were homozygotes. The analysis of dS/dN ratio (synonymous to nonsynonymous substitutions) indicated purifying selection. There were no significant associations between haplotype nor presence of particular alleles and susceptibility to infections with blood bacteria.

RITseq screening for drug targets in the protein kinome of *Trypanosoma brucei.* (P113)*

Fernando Fernandez-Cortes, Tiago D. Serafim, Nathaniel G. Jones, Ryan Ritchie, Jonathan Wilkes, Richard McCulloch, Jeremy C. Mottram.
Wellcome Trust Centre for Molecular Parasitology. Sir Graeme Davies Building University of Glasgow 120 University Place Glasgow G12 8TA

Protein kinases (PKs) are the main family of signaling proteins in eukaryotes. An *in vitro* RNA interference (RNAi) screen knocking down 190 PKs predicted for *Trypanosoma brucei*, was recently published in our lab identifying 42 important for normal cell growth and 2 involved in differentiation pathways. We now present a study where this collection of 183 individual *T. brucei* cell lines was pooled and RNAi target sequencing (RITseq) technology used to investigate the essentiality of the PKs *in vivo*. 24h after inoculation in CD-1 out bred mice, RNAi was induced for 48h (in parallel to non-induced controls), genomic DNA was isolated from the blood recovered parasites and RITseq performed using the Ion Proton® platform. As a result, 45 RNAi-induced lines were identified with a significant loss of fitness, 9 of them showing essentiality *in vivo* that was previously not observed *in vitro*. From these 9, we identified 2 PKs related with DNA repair mechanisms. The approach reveals high reproducibility over biological and technical replicates that was also observed over consecutive sequencing runs. Suitable for *in vivo* and *in vitro* investigation of any aspect of the parasite biology, this technique rises as a powerful, versatile and relatively unexpensive tool for high component screening and pathway dissection of the protein kinome in *T. brucei*, the etiological agent of sleeping sickness.

Functional characterization of the *Leishmania*-specific chaperone HSP70r (P114)*

Drini Sima, Olivier Leclercq, Gerald F. Späth
Institut Pasteur and CNRS URA2581, Unité de Parasitologie Moléculaire et Signalisation, Paris, France

A 2D-DiGE proteomics analysis of transgenic parasites overexpressing GFP-tagged MAPK7 (GFPK7) showed over-phosphorylation of a highly parasite-specific chaperone, which we termed HSP70r as it contains an HSP70-related domain and a TPR domain involved in protein-protein interaction. We applied a multi-disciplinary strategy to characterize HSP70r through bioinformatics, cell-biological, biochemical and genetics approaches. Utilizing bioinformatic approaches, including multiple sequence alignment and phylogenetic analysis, we showed that HSP70r is highly

conserved across all major *Leishmania* species, as well as in ancestral non-pathogenic trypanosomatids, but absent in related pathogenic *Trypanosma brucei* and *T. cruzi*. Compared to canonical HSP70 family members, the HSP70 domain of HSP70r is very divergent, which raises the question whether it carries ATPase function or acts as a chaperone. In order to test the capacity of recombinant HSP70r to bind to ATP affinity columns, we have cloned a GST-tagged HSP70r and purified the protein from recombinant bacteria by FPLC. Preliminary data suggest that HSP70r may not bind to ATP. We generated transgenic parasites over-expressing GFP-HSP70r in order to study its localization and reveal whether it is interacting with HSPs, co-chaperones, and so-called "client proteins". To this end, a co-immunoprecipitation using anti-GFP antibody followed by mass spectrometry analysis has been performed. Several HSP70-associated proteins have been identified, including other *Leishmania major* HSP70s (LmjF.28.2770, LmjF.30.2550, LmjF.30.2490, LmjF.30.2470) and HSP90 (LmjF.33.0330). The presence of HSP70r-containing protein complexes will further be studied using Blue Native electrophoresis and Western blot analysis. Data on protein expression, ATP-binding capacity and stage-specific interaction of HSP70r will be discussed.

Stage-specific activity of the *Leishmania* MAP kinase MPK10 is regulated by a parasite-specific auto-inhibitory domain (P115)*

Mathieu Cayla1, Najma Rachidi1, Olivier Leclercq1, Dirk Schmidt-Arras1, Heidi Rosenqvist2,3, Martin Wiese2, Gerald F. Späth1

1 Institut Pasteur and Centre National de la Recherche Scientifique URA 2581, Unité de Parasitologie Moléculaire et Signalisation, 25 rue du Dr Roux, 75015 Paris, France; 2 Strathclyde Institute of Pharmacy and Biomedical Sciences, University of Strathclyde, 161 Cathedral Street, Glasgow, Scotland; 3 Protein Research Group, Department of Biochemistry and Molecular Biology, University of Southern Denmark, Odense, Denmark G4 0RE, UK

Protozoan pathogens of the genus *Leishmania* have evolved unique signalling mechanisms that can sense changes in the host environment and trigger adaptive stage differentiation essential for intracellular infection. Only little is known on the parasite-specific protein kinase biology that underlies development of the disease-causing amastigote stage, even though protein phosphorylation has been identified as a major target for anti-leishmanial chemotherapy. Here, we unravel highly unusual regulatory mechanisms for the conserved *Leishmania* MAP kinase homolog MPK10. MAP kinases are activated by higher order MAP kinase kinases through phosphorylation on threonine and tyrosine residues inside the highly conserved TxY motif of the activation loop. Using a transgenic approach we demonstrate that the stage-specific increase in MPK10 activity during the pro- to axenic amastigote conversion does not correlate with the largely constitutive phosphorylation status of the regulatory tyrosine residue inside the kinase activation loop despite its major role for kinase activity. This suggests that MPK10 is maintained in a partially activated state and controlled by non-classical means. In fact, deletion of the last 46 amino acids of MPK10 resulted in significantly enhanced kinase activity in MPK10, thus revealing its regulation by an auto-inhibitory mechanism. Phosphoproteomics analyses identified a novel regulatory phospho-serine residue inside the C-terminal auto-inhibitory domain at position 395 that is likely implicated in MPK10 regulation. Together our data uncover highly unusual and novel aspects of parasite-specific protein kinase regulation, and propose MPK10 as a potential signal sensor of the mammalian host environment, whose intrinsic pre-activated conformation is regulated by auto-inhibition.

Under-reporting human trypanosomiasis outbreak in endemic districts in central Uganda (P116)*

Acup Christine Amongi1,2, Kakembo Abbas Luberenga3, Picozzi Kim1, Waiswa Charles4, Kabasa John David2, Ian Maudlin1, Sue Welburn1.

1 Centre for Infectious Diseases, University of Edinburgh, EH16 4SB, Scotland, UK; 2 College of Veterinary Medicine Animal Resources and Biosecurity, Makerere University, Kampala, Uganda; 3 Vector Control Division, Ministry of Health, Kampala, Uganda; 4 Control of Trypanosomiasis in Uganda, Kampala

Currently, classifying the type of sleeping sickness infection at the point-of-care in Uganda is dependent on geographical location of the patient. HAT due to *T. b. gambiense* is chronic, quiescent in the Northwest of the country and an estimated two million people are at risk, while an estimated nine million people are at risk for HAT due to *T. b. rhodesiense* that is endemic in the areas around the Lake Basin of Lake Victoria and Lake Kyoga. Uganda is the only country where both *T. b. gambiense* and *T. b. rhodesiense* HAT occur within a single national border. Although both diseases are frequently considered together, they are clinically, epidemiologically and geographically distinct. The 2009 epidemic of human trypanosomiasis in Soroti, Kaberamaido and Dokolo Districts, provided an opportunity to assess the level of under detection (and reporting) of disease outbreaks from within communities reporting with disease to sleeping sickness hospitals. Here we present an analysis of under detection of disease in both active and passive surveillance

following an epidemic in the aforementioned endemic districts. In summary, our assessment shows that active surveillance detects more cases than passive surveillance, and that recognition (and reporting) in passive surveillance is reduced by inefficiencies and deficiencies in the healthcare delivery system.

Differential exposure to sandfly bites in cutaneous leishmaniasis patients and healthy individuals from different endemic areas in Saudi Arabia (P117)

Karina Mondragon-Shem1,2, Waleed S Al Salem1,2,3, Mohamed Alzahrani3, Ziad Memish3, Abdelmohsin M Abdon3, Maha Abdeladhim4, Jesus G. Valenzuela4, Alvaro Acosta-Serrano1,2.
1. Parasitology Department, Liverpool School of Tropical Medicine, Pembroke Place, L3 5QA, UK. 2. Vector Department, Liverpool School of Tropical Medicine, Pembroke Place, L3 5QA, UK. 3. Saudi Ministry of Health, Riyadh, Kingdom of Saudi Arabia. 4. Vector Molecular Biology Section, Laboratory of Malaria and Vector Research, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Rockville, Maryland, United States of America.

Antibody levels against vector salivary proteins indicate exposure to bites, which has been correlated with risk of disease development. *Leishmania major* is the main causative agent of cutaneous leishmaniasis (CL) in the Kingdom of Saudi Arabia (KSA), where it is mainly transmitted by *Phlebotomus papatasi*. Salivary protein SP32 has been identified as the most immunogenic protein in *Ph. papatasi* saliva, making it useful to measure exposure to the bite of this species. In this work, we determined the level of exposure to *Ph. papatasi* bites in individuals from several CL endemic areas in KSA. We obtained sera from CL patients, CL-cured individuals and healthy individuals from Al-Hasa, Al-Madinah and Asir, and measured the levels of anti-SP32 antibodies by ELISA using a recombinant SP32 antigen. Sandflies were collected from the same regions in order to identify species. We found the titres of anti-SP32 antibodies were higher in CL patients than healthy and CL-cured individuals in Al-Hasa, while in Al-Madinah there was no significant difference between the same groups. Interestingly, we found a correlation between high levels of anti-SP32 antibodies and nodular lesion development in patients from Al-Madinah. Entomological analyses found *Ph. papatasi* was the dominant species in both Al-Hasa and Al-Madinah, while *Ph. sergenti* (*L. tropica* vector) was more common in Asir where only background levels of anti-SP32 antibodies were found. This is the first study using recombinant SP32 protein to assess biting exposure in CL patients. Our results suggest a correlation between biting exposure, disease outcome and clinical presentation.

A *Trypanosoma brucei* whole genome RITseq screen reveals novel protein kinases involved in DNA repair (P118)*

Tiago Donatelli Serafim1, Jennifer Ann-Black1, Jon Wilkes1, Sam Alsford2, David Horn3, Jeremy C. Mottram1 and Richard McCulloch1
1 Wellcome Trust Centre for Molecular Parasitology and Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, United Kingdom. 2 London School of Hygiene & Tropical Medicine, London, United Kingdom. 3 Biological Chemistry & Drug Discovery, College of Life Sciences, University of Dundee, Dundee, UK.

Protein kinases (PKs) are important for many signaling pathways in eukaryotes. This work investigated PKs in the African trypanosome *Trypanosoma brucei* that contribute to the control or execution of DNA repair, fundamental pathways to ensure fidelity and transmission of the genetic material. To date, virtually no PKs that act in repair have been described in any kinetoplastid, nor considered for their potential in therapeutic intervention. A whole-genome RNAi screen, adapted from previously detailed 'RITseq', was performed in bloodstream *T. brucei* cells: RNAi was induced for 24 hrs and the cells then grown for a further 4 days in the presence or absence of MMS, an alkylating agent that causes DNA breaks and replication stalling. Genomic DNA was isolated and analyzed by next generation sequencing, and loss of fitness in the presence of MMS was identified for 14 PKs due to reduced sequence reads relative to the untreated controls. To validate these candidates, cells for inducible RNAi against each were generated and examined for growth, cell cycle progression and gamma-H2A accumulation after MMS treatment. 7 PKs were confirmed to have roles in DNA repair or associated processes. For only one, TLK, has a DNA repair function been suggested (in yeast), meaning that all others represent novel DNA repair PKs. Further validation of PK function in DNA repair will involve a kinome-wide RITseq in *T. brucei*, as well as detailed dissection of the role of the each of the 7 PKs in this crucial genome maintenance pathway.

Evolution of membrane trafficking in kinetoplastids (P119)*

Divya Venkatesh1, Amanda J. O'Reilly1, Paul T. Manna1, Steve Kelly2, Mark C. Field1
1 Division of Biological Chemistry and Drug Discovery, University of Dundee, Dow Street, Dundee, Scotland, DD1 5EH, 2

Oxford Centre for Integrative Systems Biology, Department of Biochemistry, University of Oxford, South Parks Road, Oxford OX1 3QU, UK

The basic organisation and molecular machinery of membrane trafficking is an ancient and conserved eukaryotic feature. We are exploiting newly available kinetoplastid (trypanosome) genome sequence resources to enquire how intracellular trafficking varies across these organisms and how this correlates with life cycle, virulence and taxonomy. We chose to study Rab and SNARE gene products, which encode much of the specificity for this system. Evolution within these gene families can provide insight into the atlas of the endomembrane system. The SNARE and Rab repertoires were established prior to divergence of the kinetoplastids and both families are relatively stable. However, significant smaller scale changes are evident, including the presence of possible kinetoplastid-specific paralogs and lineage-specific expansions and deletions. In parallel, we epitope-tagged all predicted R-SNAREs in *Trypanosoma brucei* for both visualisation and as affinity handles for isolation of SNARE complexes. Our data define the specificity within the endomembrane system of kinetoplastids and identify likely adaptations accompanying selection and virulence.

Introducing Leishmaniac.org, a molecular parasitology resource (P120)

Pegine Walrad
Centre for Immunology and Infection, Department of Biology, University of York

The website http://www.Leishmaniac.org is a new protocol resource for cell and molecular biologists working with *Leishmania* spp. parasites. Protocols, methods and molecular markers can differ significantly within the Leishmaniasis research field. Combined with the diversity of parasite species under investigation, this complicates direct data comparisons and can obstruct progress. Leishmaniac.org is designed to help overcome this by encouraging protocol sharing, data communication and constructive discussion within the *Leishmania* spp. research community. This site complements but does not replicate other Leishmaniasis web resources and databases such as leishnet.net, tritrypdb.org, and genedb.org. The inclusive aim is to promote, encourage and facilitate round the clock information and technique sharing between international researchers in our field. It is designed as an information hub to assist the training of both students and transitioning molecular biologists. This will be the first unveiling of the website and a brief demonstration will be given. Leishmaniac.org resources, links and format will be presented.

Leishmania* virulence factors: Inhibitors of serine peptidases (P121)

Amy Goundry1,, Elmarie Myburgh1, Ana Paula C. A. Lima2 and Jeremy C. Mottram1
1 Wellcome Trust Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, Glasgow, G12 8TA, UK, 2 Instituto de Biofísica Carlos Chagas Filho, Universidade Federal do Rio de Janeiro, Rio de Janeiro, RJ, 21949-900, Brazil

Sequencing of the *Leishmania major* genome has revealed the presence of three ecotin-like peptide inhibitors of trypsin-family serine peptidases (ISPs). As serine peptidases are absent in *Leishmania*, these ISPs have been proposed to inhibit host serine peptidases, with ISP2 expressed in the mammalian-infective metacyclic promastigote and amastigote stages. ISP2 has previously been shown to inhibit neutrophil elastase (NE), a serine peptidase expressed by mammalian innate immune cells, including neutrophils, monocytes, and macrophages. This inhibition prevents a Toll-like receptor 4 (TLR4)-NE pathway during *Leishmania*-macrophage interaction promoting *Leishmania* growth in macrophages *in vitro*. Using *L. major* wild-type and an ISP2/3 knock-out (Δisp2/3) mutant, the molecular basis for ISP2 as a virulence factor is currently being investigated *in vivo* by examining interactions with innate cells and tracking disease progression. Quantitative *in vivo* bioluminescence imaging of myeloperoxidase activity of activated phagocytes has been determined over a period of 7 weeks. The recruitment of innate cell populations and iNOS expression at the site of inoculation during the course of infection have also been examined by flow cytometry. These studies have revealed differences between *L. major* wild-type and Δisp2/3 infection coinciding with a second wave of innate cell infiltration. The use of transgenic mice deficient in NE has enabled an investigation into the role of this host serine peptidase during *in vivo* infection. We will discuss the effect of ISP2 on the innate immune response, innate cell recruitment and activation and how this contributes to disease progression during *L. major* infection.

From the laboratory to the real world: wild rodents as a genetic model of disease susceptibility (P122)

Andrew Turner, Steve Paterson
Institute of Integrative Biology, Biosciences Building, University of Liverpool, Crown Street, Liverpool L69 7ZB.

Individuals vary in their susceptibility to infectious disease, and it is now well established that host genetic factors form a major component of this variation. The discovery of genes underlying susceptibility has the potential to lead to improved disease control through the identification and management of vulnerable individuals and the discovery of novel therapeutic targets. Whilst laboratory rodents have proved invaluable for elucidating the functions of genes involved in immunity to infection, these captive animals experience conditions very different to the natural environment, lacking the genetic diversity and environmental pressures characteristic of natural populations, including those of humans. It has therefore often proved difficult to translate basic laboratory research to the real world. In order to further our understanding of the genetic basis of infectious disease resistance, and the evolutionary forces that drive variation in susceptibility, we propose that genetic research traditionally conducted on laboratory animals is expanded to the more ecologically valid arena of natural populations. Here we briefly highlight the potential of using wild rodents as a new resource for biomedical research, to link the functional genetic knowledge gained from laboratory rodents with the variation in infectious disease susceptibility observed in humans and other natural populations.

A simple, rapid and scalable bioluminescence assay to estimate antimalarial rate of kill *in vitro* (P123)*

Imran Ullah, Rhiannon Blow and Paul Horrocks
Institute for Science and Technology in Medicine, Keele University, Staffordshire ST5 5BG, United Kingdom

Fast-acting antimalarial drugs save lives and reduce the morbidity of disease. With tens of thousands of new drugs in the discovery pipeline, there is an urgent need to establish scalable high-throughput assays to prioritise targets for development. Here we present a novel bioluminescence-based assay used to estimate the rate-of-kill of antimalarial drugs *in vitro* – using a simple, rapid and readily scalable format. We provide data that validates the assay against benchmark antimalarial drugs, extending this to show results from a an ongoing screen of leading development targets available through the Medicines for Malaria Venture's Malaria box.

Antiparasitic pyrrolopyrimidines (P124)

Federica Giordani1, Colin J. Suckling2, Colin L. Gibson2, Judith K. Huggan2, Abedawn I. Khalaf2, William N. Hunter3, Keri L. Barrack3, Michael P. Barrett1
1 Wellcome Trust Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, Sir Graeme Davis Building, University of Glasgow, Glasgow, G12 8TA UK, 2 WestCHEM, Department of Pure and Applied Chemistry, University of Strathclyde, Glasgow, G1 1XL UK 3 Division of Bilogical Chemistry and Drug Discovery, College of Life Sciences, University of Dundee, Dundee, DD1 5EH UK.

Human African trypanosomiasis (HAT) is a vector-borne disease caused by two subspecies of *Trypanosoma brucei* protozoan parasites. HAT belongs to the so-called "neglected tropical diseases", mainly affecting poor communities in Sub-Saharan Africa. Despite the significant drop in reported cases over the last decades, and the progression through clinical trials of a couple of new compounds, the efforts to fight this disease must not be abandoned, to avoid resurgence. Current chemotherapy, which has a key role in control programs, is still highly unsatisfactory, and improved drugs are required. Our quest for more effective treatments to use against HAT led to the synthesis of a series of pyrrolopyrimidine compounds targeting the pteridine reductase (PTR1) enzyme, previously shown to be essential in trypanosomes. PTR1 is unique to trypanosomatids, where it reduces biopterins to their biologically active form, and where it also provides an alternative route to reduced folates to the normal dihydrofolate reductase. The pyrrolopyrimidine design, guided by crystallography and activity data, allowed us to generate molecules with good enzymatic inhibition profiles and *in vitro* activity in the high nanomolar range against *Trypanosoma brucei brucei*. Parasites treated with these molecules presented with cell division defects. Despite the targeted approach adopted, metabolomics results and inhibitory curves indicated possible multiple mechanism of action. *In vivo* experiments gave some evidence of parasite clearance, but they could not be completed due to toxicity issues.

Trypanosomiasis in working equids in West Africa: characterising neurological disease (P125)*

Demelza Kingston, J. Rodgers, S. Sharpe, P. Capewell, W. Weir, B. Bradley, D.G.M. Sutton
Institute of Biodiversity, Animal Health and Comparative Medicine, School of Veterinary Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, UK

Rural populations in The Gambia are dependent on working equids for socioeconomic development. Multiple

trypanosome variants including *Trypanosoma congolense, T. vivax* and the *T. brucei* subspecies (*T. b. brucei, T. b. evansi and T. b. equiperdum*) are prevalent in the region. A causal link has been established between a multifocal neurological syndrome of equids and CNS trypanosome infestation. No effective treatment is available and the disease is generally fatal. Equine CNS trypanosomiasis is of international importance with cases reported in Mali, Tanzania, Ethiopia, Venezuela and India. Six patients presented to Gambia Horse and Donkey Trust, a charity providing veterinary care to working equids, with clinical signs consistent with CNS trypanosomiasis were included in this study. In horses, progressive spinal ataxia predominated. Affected donkeys more commonly displayed behavioural abnormalities and cranial nerve deficits with mild spinal ataxia. All patients deteriorated and were euthanized on humane grounds. In all cases, histopathology showed diffuse lymphocytic-plasmacytic meningoencephalomyelitis with marked perivascular cuffing. Pathology was most severe in the white matter. Immunohistochemistry identified *Trypanosoma brucei* subspecies within CNS tissue sections. Where appropriate samples were available, *Trypanosoma brucei* DNA was also detected within CNS tissue (4/4 cases) and blood (2/4 cases) by standard PCR. Microsatellite markers were used to genotype CNS and circulating parasites. Results from 3 markers (Ch5/JS2, Ch11/110 and Ch11/51) indicate the presence of at least 4 genotypes within the study population. Additional microsatellites will be employed to consolidate these findings. Sample collection is ongoing.

A model for leptospire dynamics in its reservoir host in a favela setting (P126)

Amanda Minter, Mike Begon, Jamie Childs, Federico Costa, Peter Diggle, Albert Ko
University of Liverpool

Leptospirosis is a zoonosis that humans can contract via contact with animal reservoirs directly or with water contaminated with their urine. Salvador, Brazil has had a recent population increase which has led to the creation of urban slums, ('favelas') which are overcrowded and lack basic sanitation. The conditions of the favelas favour rodent borne transmission of leptospirosis. The Norway rat (*Rattus norvegicus*) is asymptomatic and can transmit the disease for the entirety of its life. By identifying and quantifying the ecological factors driving leptospire dynamics in its reservoir host in tropical slums, patterns of human infection can be predicted. A simple mechanistic model is presented for leptospire dynamics in its reservoir host consisting of three ordinary differential equations: for susceptible and infected rats and for free living leptospires in the environment. Ongoing studies to parameterize the model are also described. This will be the first study to examine leptospire dynamics in its reservoir host in a favela setting.

Major genetic risk factor for visceral leishmaniasis provides new prospects for understanding the host immune responses (P127)

Michaela Fakiola1, Amy Strange2, Heather J Cordell3, E Nancy Miller4, Mary E Wilson5, Selma M Jeronimo6, Shyam Sundar7, Chris CA Spencer2, Peter Donnelly2,8, Jenefer M Blackwell4,9, The LeishGEN Consortium and the WTCCC2
1 Department of Pathology, University of Cambridge, Tennis Court Road, Cambridge, UK, 2 Wellcome Trust Centre for Human Genetics, University of Oxford, Oxford, UK, 3 Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK, 4 Cambridge Institute for Medical Research, University of Cambridge School of Clinical Medicine, Cambridge CB2 0XY, UK, 5 Departments of Internal Medicine and Microbiology, University of Iowa and the VA Medical Center, Iowa City, IA, USA 52242, USA, 6 Department of Biochemistry, Center for Biosciences, Universidade Federal do Rio Grande do Norte, Natal, RN 59078-970, Brazil, 7 Institute of Medical Sciences, Banaras Hindu University, Varanasi – 221 005, India, 8 Department of Statistics, University of Oxford, Oxford, UK, 9 Telethon Institute for Child Health Research, Centre for Child Health Research, The University of Western Australia, Subiaco, Western Australia 6008, Australia

The clinical outcome of infection to *Leishmania* parasites is influenced by the complex interaction between parasite strain, host genetics and environmental factors. The importance of host genetic factors in human susceptibility to visceral leishmaniasis (VL) is indicated by familial clustering and high sibling risk ratios, however human genetic studies undertaken to date have largely been underpowered. In collaboration with the Wellcome Trust Case Control Consortium 2 we recently reported the first genome-wide association study (GWAS) of VL across major foci of disease caused by *L. donovani* in India and *L. infantum chagasi* in Brazil. The HLA-DRB1–HLA-DQA1 locus within the Major Histocompatibility Complex (MHC) was highlighted as the single most important genetic determinant of VL susceptibility (Pcombined=2.76x10^17; odds ratio=1.41; 95% confidence interval=1.30-1.52 over three cohorts). We further showed that specific HLA-DRB1 allele groups are correlated with disease risk (DRB1*14/*13/*11) versus protection (DRB1*15/*16/*01), a correlation that remarkably crosses the geographical and epidemiological divides of continent and aetiological parasite species. In light of the role of HLA-DRB1 molecules in epitope selection and antigen presentation, the GWAS findings go to the heart of eliciting T cell immunity against *Leishmania* parasites. By employing a combination

of *in silico*, *in vitro*, and *in vivo* methods we can address questions regarding the repertoire of pathogen peptides presented by the specific risk versus protective HLA-DRB1 alleles. This knowledge can provide important leads in understanding the host-pathogen interactions and T-cell mediated responses that determine outcome of infection.

Plasmodium* and *Trypanosoma brucei* GPR89 Homologues (P128)

Rachel Milne, Keith R. Matthews, Joanne Thompson
Institute of Immunology and Infection Research/Centre for Immunity, Infection and Immunology, University of Edinburgh

The protozoan parasites *Plasmodium* and *Trypanosoma brucei* each have complex lifecycles in which they must adapt to the diverse environments of their insect vector and mammalian host. The signalling mechanisms involved in their differentiation between different life forms are largely unknown. Interestingly, both parasites possess molecules of the phylogenetically widespread GPR89 family of putative Receptors or Channels that have been implicated in G protein signalling in plants and Golgi acidification in mammals. Here we set out to analyse the *P. chabaudi* and *T. brucei* GPR89-like proteins biochemically and functionally to elucidate their potential role in environmental sensing. Moreover, by expressing these molecules as recombinant proteins or in cell based reporter systems, we are developing screening assays to identify chemical inhibitors or activators of the proteins. Our functional analyses of the role of these molecules in *T. brucei* and *P. chabaudi* suggests that pharmacological intervention in their function is likely to have important consequences for parasite virulence or transmission.

New insights into the role of TbCentrin 2 in basal body and flagellum biogenesis (P129)*

Sam Barry, Katie Towers, Sue Vaughan
Oxford Brookes University, Faculty of Health and Life Sciences, Department of Biological and Medical Sciences, Gipsy Lane, Headington, Oxford, OX3 0BP

The flagellum of *Trypanosoma brucei* is essential for pathogenicity and is assembled from a microtubule-based cylindrical structure called a basal body (analogous to centrioles in mammalian cells). During the cell division cycle a new flagellum is assembled alongside the old flagellum and assembly requires duplication and segregation of basal bodies. Basal bodies exist as a pair with a defined 'age'. The mature basal body can extend a flagellum but the pro-basal body, which is positioned alongside cannot extend a flagellum until the cell cycle after it was assembled. During the next cell division cycle the pro-basal body matures and can therefore assemble a flagellum and two new pro-basal bodies form alongside each mature basal body. Centrin is a calcium-binding protein which has been localised to a number of cytoskeletal structures including the basal body and flagellum in eukaryotic organisms. There are 5 centrin proteins in *Trypanosoma brucei*. Centrin 2 (TbCen2) has previously been localised to the basal bodies, bilobe and flagellum. Localisation studies of 20H5 which recognises Centrin 1 and 2 in *T. brucei* revealed a distinct pattern of labelling corresponding to the 'age' of each basal body. This pattern of labelling was also demonstrated by localisation studies of endogenous YFP:TbCen2 fusion in addition to localisation at the bilobe and flagellum. We also discovered defects in the assembly of central pair microtubules of the axoneme following knockdown of TbCen2 by RNAi. These results give further insights into the multi-functional role of this family of proteins in eukaryotic cells.

Lectin Pathway inhibition by parasitic scabies mites: molecular characterisation of host-pathogen immune mechanisms (P130)

Simone Reynolds1, Angela Mika2, Robert Pike3, Anna Blom4, Dave Kemp1, Katja Fischer1
SReynolds,DKemp, KFischer - 1 QIMR Berghofer Medical Research Institute, Brisbane, Australia; 2 Bernhard Nocht Institute for Tropical Medicine, Hamburg, Germany; 3 Monash University, Melbourne, Australia; 4 Lund University, Malmö, Sweden

Scabies is a parasitic skin infestation caused by the mite *Sarcoptes scabiei*. Common worldwide, it remains a major public health problem in socially disadvantaged populations, including Australian Indigenous communities. Scabies lesions are commonly co-infected with opportunistic bacteria, particularly Group A streptococci (GAS). In Australian Indigenous communities the increased streptococcal burden has been associated with extreme levels of Rheumatic Heart Disease and Rheumatic Fever. Mass scabies treatment programs have resulted in reduced scabies prevalence coinciding with significantly reduced rates of bacterial infections. Hence scabies eradication may result in improved health outcomes. Recent reports of mite resistance to current scabicides, emphasises the need to identify novel targets for protective intervention. We have identified two scabies mite protease paralogues which facilitate host immune evasion by

inhibiting the lectin pathway. These two proteases are members of a multigene family that are catalytically inactive due to structural restraints and mutations in the catalytic triad. They have been named Scabies Mite Inactive Serine Protease Paralogues (SMIPP-Ss). Employing site-directed mutagenesis we are identifying the complement binding site(s) in the two SMIPP-Ss to determine the binding mechanism. Initial studies suggest inhibition is through binding to the collagen region of MBL. Studies are currently determining if MASP displacement is also a factor. This research may lead to the development of novel parasitic therapeutics or the identification of anti-complement compounds suitable for complement-associated disease therapy.

A whole-cell, target-based approach to antimalarial drug discovery (P131)

Catherine Moore, Hatoon Niyazi, Henry M. Staines, Sanjeev Krishna
Centre for Diagnostics and Antimicrobial Resistance, Infection and Immunity Research Institute, St. George's University of London

Over the past few decades there has been an arms race between our attempts to control malaria, and the parasites developing resistance to almost all available drugs. Antimalarial drug discovery is, therefore, of paramount importance. Target-based drug screening is an advantageous system for drug discovery because it both identifies new inhibitors and also provides information on the mechanism of action. However, *in vitro* assays on integral membrane proteins, which are often effective drug targets, are difficult due to protein solubility and folding problems. To circumvent this, heterologous expression systems are used to allow screening of small molecule inhibitors. The calcium pump PfATP6 is a validated drug target and there is a great deal of evidence to suggest this pump is involved in the artemisinins' mode of action. We have expressed this pump in a strain of *Saccharomyces cerevisiae* which has had the endogenous calcium and drug efflux pumps deleted. Yeast growth is rescued in the presence of calcium by the expression of PfATP6. For the screening assay, yeast are incubated in the presence of the screening compounds and the growth is measured (by absorbance at 620 nm). To determine the compounds' inhibitory effect, results are compared with no drug controls. Before this, however, the assay needs to be optimised in terms of sensitivity and calcium tolerance (in both PfATP6 and control strains, which express either the mammalian ortholog or the vector only). Here, we present the development of this system.

Exo99 is a novel component of exocyst in *Trypanosoma brucei* (P132)

Cordula Boehm1, Samson Obado2, Michael Rout2, Brian Chait2, Mark Field1
1 Division of Biological Chemistry and Drug Discovery, University of Dundee, Dow Street, Dundee, Scotland, DD1 5EH, 2 The Rockefeller University, New York, NY10021,USA

In trypanosomatids post-Golgi trafficking and the secretory pathway is less well characterised than the endocytic system. Exocyst, a multimeric complex, targets post-Golgi vesicles to the plasma membrane where they fuse via SNARE interactions. In higher eukarya exocyst is an octameric complex that consist of two subcomplexes of three and five subunits each, by comparative genomic and phylogenetic analysis only 6 out of 8 subunits were identified in trypanosomatids. With a novel cryo grinding affinity purification approach we have purified exocyst from *Trypanosoma brucei* using Sec15 as a bait. With this approach we could not only purify orthologs of all 8 exocyst subunits from *Saccharomyces cerevisiae* but also discovered a novel subunit we have named Exo99. Reverse pullouts using Exo99 as the bait isolated the entire exocyst complex and confirmed Exo99 as a exocyst component in trypanosomatids. Exo99 an essential component in *Trypanosoma brucei* is restricted to kinetoplastids, its precise function in exocytosis remains to be elucidated.

Insights on immunological and histopathological features of *L. infantum*/*L. major* hybrid strains in BALB/c mice infection (P133)*

Andreia Albuquerque1, Sofia Cortes1,2, Carla Maia1,2, Maria Carvalho3, Luiz AR de Freitas4, Washington LC dos-Santos4, Lenea Campino1,5
1 Instituto de Higiene e Medicina Tropical (IHMT), Universidade Nova de Lisboa (UNL), Lisboa, Portugal; 2 Centro de Malária e outras Doenças Tropicais, IHMT, UNL, Lisboa Portugal; 3 Laboratório de Anatomia Patológica, Clinilab, Grupo Galenus, Lisboa, Portugal; 4 Centro de Pesquisas Gonçalo Moniz, Fiocruz, Salvador da Baía, Brazil; 5 Departamento de Ciências Biomédicas e Medicina, Universidade do Algarve, Faro, Portugal

Leishmaniasis are worldwide diseases caused by protozoan parasites of *Leishmania* genus. Balance of immune response

is the key factor that regulates resistance or progression to active infection. Histopathology allows contrasting the main tecidual alterations, as inflammation and cellular response. Previous studies found that New World hybrid strains were more resilient than their putative parents. This study evaluated the presence/absence of parasites, expression of pro and anti-inflammatory cytokines and histopathological features in different tissues of BALB/c mice infected with *L. infantum/L. major* (Old World) hybrids during an eight week course of infection. The results showed that both infections by hybrid and putative parental strains seem to have a mixed Th1/Th2 immune response. On the other hand, despite the recovery of parasites in all time-points and important histological alterations observed, the cytokines' expression by hybrids was detected later than in parental strains' infection, suggesting a higher capacity of "camouflage" by these hybrids regarding the activation of the immune system in comparison to their parental strains. Occurrence and emergence of *Leishmania* hybrid strains, along with its virulence and dynamics of transmission are fairly studied and are imperative for disease control. To our knowledge this is the first report concerning immunological and histopathological features of *L. infantum/L. major* hybrids *in vivo* infection.

Anti galactosyl α(1-3)gal antibodies in patients with Old World cutaneous leishmaniasis: a potential marker for clinical features and disease cure (P134)

Waleed S Al Salem1,2, Daniela Ferreira1, Naomi Dyer1, Salah M Balghonaim2, Ahmed Y Al Muhanaa2, Mohamed H Alzahrani2,Ali Al Shahrani2, Mohammed A. Aldahan2 ,Abdulaziz Al Jarallh2 ,Saleem Al-Zubiany2, El-Keir Ibrahim2, Ziad Memish2, Igor C. Almeida3, Alvaro Acosta- Serrano1

1 Department of Parasitology, Liverpool School of Tropical Medicine, UK, 2 Saudi Ministry of Health, Riyadh, Saudi Arabia, 3 Department of Biological Sciences, University of Texas at El Paso, USA.

Anti-alpha galactosyl (anti-αGal) antibodies represent ~1% of total human IgG as a result of gut microbiota expressing αGal epitopes. In infections with *T. cruzi* and several species of New World *Leishmania* parasites, high levels of anti-αGal have been reported. However, nothing is known about the levels of anti-αGal in Old World cutaneous leishmaniasis (CL) patients or its relationship with pathology. Here, we measured the serum levels of anti-αGal antibodies from CL-infected individuals (either infected with *Leishmania major* or *L. tropica*) from different endemic regions from Saudi Arabia. To determine anti-αGal titers, we developed a novel chemiluminescent ELISA assay that uses the synthetic neoglycoprotein antigen (NGA), Galα1-3Galβ1-4GlcNAc-BSA. Using this assay, we have found that individuals infected with either *Leishmania* spp have significantly elevated titres of Leish-αGal antibodies compared to healthy individuals, also from CL endemic regions. Interestingly, cured CL individuals (up to 2 years) contain six- to eight-fold levels of anti-αGal antibodies compared to healthy volunteers. Moreover, the levels of αGal IgG increase with the number of lesions and were found higher in patients with nodular or ulcerated nodular lesions. Furthermore, when separated by subclasses, IgG1 and IgG3 were found significantly higher during an active infection, while IgG2 and IgG4 are higher during re-epithelialization. Preliminary screenings of a library of novel NGAs, containing different αGal terminal epitopes, using sera from either *L. major* or *L. tropica*, allows the identification of glycotopes that can discriminate between the infections with either parasite species.

A genetic backcrossing approach to identify genetic loci contributing to ivermectin resistance in *Haemonchus contortus* (P135)

Axel Martinelli1, Elizabeth Redman2, Roz Laing3, Dave Bartley4, Alan Tracey1, Karen Brooks1, Nancy Holroyd1, Matthew Berriman1, Chris Illingworth5, James Cotton1, John Gilleard2

1 Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SA, UK, 2 Deptment of Comparative Biology and Experimental Medicine, Faculty of Veterinary Medicine, University of Calgary, TRW 2D10, 3280 Hospital Drive NW, Calgary, Alberta, Canada T2N 4Z6, 3 Institute of Infection, Immunity and Inflammation, Henry Wellcome Building, School of Veterinary Medicine, Bearsden Road, University of Glasgow, Glasgow, G61 1QH, 4 The Moredun Group, Pentlands Science Park, Bush Loan, Penicuik EH26 0PZ, Scotland, UK 5 Department of Genetics, University of Cambridge, Downing Street, Cambridge CB2 3EH, UK.

Haemonchus contortus is an important sheep parasite, as well as a model used in the study of both the mechanistic and genetic basis of drug resistance in helminths. Two independent fourth-generation genetic back-crosses between a fully ivermectin sensitive MHoc3 (ISE) and two geographically distinct drug resistant strains, namely MHoc10 (CAVR) and MHco4 (WRS), have been previously generated. The aim of these crosses was to introgress regions of the genome harbouring drug resistance loci from the resistant parental strain into the genetic background of the susceptible ISE strain to ultimately allow the identification of genes involved in drug resistance. We present current progress on a genome-wide analysis of these back-crosses in which we identify scaffolds from the current ISE genome assembly that

reside within the introgressed regions. Ongoing work on the assembly of the reference genome should progressively improve the resolution of the results. This information will be coupled with a number of improved statistical approaches to analysing back-cross experiments in small populations to allow more accurate identification of loci associated with IVR resistance and eventually identify candidate genes for functional follow-up studies.

The R enantiomer of the anti-tubercular drug PA-824 as a potential oral treatment for visceral leishmaniasis (P136)

Susan Wyllie, Stephen Patterson, Alan Fairlamb

Division of Biological Chemistry and Drug Discovery, Wellcome Trust Biocentre, College of Life Sciences, University of Dundee, Dundee DD1 5EH, Scotland, UK.

The novel nitroimidazopyran agent, (S)-PA-824, has potent antibacterial activity against *Mycobacterium tuberculosis in vitro* and *in vivo* and is currently in Phase II clinical trials for TB. In contrast to *M. tuberculosis*, where (R)-PA-824 is inactive, we report here that both enantiomers of PA-824 show potent cidal activity against *Leishmania donovani*, the causative agent of visceral leishmaniasis (VL). In *Leishmania*-infected macrophages (R)-PA-824 is 6-fold more active than (S)-PA-824. Although the *in vitro* and *in vivo* pharmacological profiles of both enantiomers are similar, (R)-PA-824 is more efficacious in the murine model of VL, with >99% suppression of parasite burden when administered orally at 100 mg kg^{-1}, twice daily for 5 days. In *M. tuberculosis* (S)-PA-824 is a prodrug that is activated by a deazaflavin-dependent nitroreductase (Ddn), an enzyme which is absent in *Leishmania* spp. Unlike nifurtimox and fexinidazole, transgenic parasites overexpressing the *Leishmania* nitroreductase are not hypersensitive to either (R)-PA-824 or (S)-PA-824, indicating that this enzyme is not the primary target of these compounds. Drug combination studies *in vitro* indicate that fexinidazole and (R)-PA-824 are additive, whereas (S)-PA-824 and (R)-PA-824 show mild antagonistic behaviour. Thus (R)-PA-824 is a promising candidate for late lead optimisation for VL and may have potential for future use in combination therapy with fexinidazole, currently in Phase II clinical trials against VL. Here, we report our preliminary findings as we attempt to define the mechanism of action of this promising nitro drug in *L. donovani*.

Multilocus sequence and microsatellite typing *Leishmania donovani* in Eastern Sudan reveal temporal dynamics of disease transmission (P137)

Rania Baleela1,3, Martin S. Llewellyn1,4, Sinead Fitzpatrick1, Katrin Kuhls2, Gabriele Schönian2, Michael A. Miles1 , Isabel L. Mauricio1,5

1 Department of Pathogen Molecular Biology, Faculty of Infectious & Tropical Diseases, London School of Hygiene and Tropical Medicine, Keppel Street, WC1E 7HT London, UK, 2 Institut für Mikrobiologie und Hygiene, Charité Universitätsmedizin, Berlin, Germany, 3 Current address: Department of Zoology, Faculty of Science, University of Khartoum, Khartoum P.O Box 321, Sudan, 4 Current address: Molecular Ecology and Fisheries Genetics Laboratory, School of Biological Sciences, University of Wales, Bangor, Deiniol Road, Bangor, Gwynedd, LL57 2UW, UK, 5 Current address: Instituto de Higiene e Medicina Tropical/Unidade de Parasitologia e Microbiologia Médicas, UEI Parasitologia Médica, Rua da Junqueira, 100, 1349-008 Lisboa, Portugal

Visceral leishmaniasis (VL), caused by the *Leishmania donovani* complex, has been responsible for devastating VL epidemics in the Sudan. Recent work has sought to elucidate the population genetics of these parasites in the Sudan/Ethiopia focus using microsatellite analysis. Here we analysed a large sample of *L. donovani* from Sudan, including a large panel of cloned strains, by both multilocus microsatellite typing (MLMT, 14 markers) and multilocus sequence typing (MLST, 11 targets). There was strong agreement between the MLMT and MLST analyses. MLST indicated the presence of two principal groups in Sudan, as previously found by MLMT. Six subpopulations were identified by cluster analysis of MLMT data. Genotypic differences between some clones for the same strain suggested the presence of mixed strains. Analysis of cloned *L. donovani* from two populations supported predominant clonality, and no mosaic break points within MLST markers were detected, and yet FIS values were consistently high, possibly explicable by inbreeding or gene conversion. Over three different years some sympatric temporal genetic changes were observed. The occurrence of canine and human isolates with the same genetic group indicated that canids may form part of the transmission cycle of VL in Sudan, with implications for control of endemic and epidemic VL. Our findings complement, with different perspectives, other population studies of *L. donovani* in Sudan, as will be discussed.

Comparative analysis of xenoma structure in five microsporidia infecting teleost fish from tunisian coasts (P138)*

lamjed Mansour1,2, Suliman Y. Alomar1

1 Department of Zoology, College of Science, King Saud University PO. Box: 2455, Riyadh, 11451, Saudi Arabia, 2 Unité de

Recherche de Biologie Écologie et Parasitologie des Organismes Aquatiques, Département de Biologie, Faculté des Sciences de Tunis, Campus Universitaire, 2092 Tunis El-Manar, Tunis

Microsporida are fungal-like eukaryotic intracellular parasites of a large number of vertebrates and invertebrates animals. They are of veterinary and medical importance since they infect silkworm, fish and also human. Their polar tube is considered as unique in the living things. This polar tube, considered as part of their strategy for invading new host cells, allow the parasite to inject the sporoplasm in the cytoplasm. In infected fish, microsporidia induce the formation of a cyst like structure, known as xenoma. This structure is a unique infected cell, having undergone hypertrophy of its cytoplasm and its nucleus, allowing the development and multiplication of the parasite. We present in this work a comparative analysis of different xenoma interfaces developed by the parasite in fish infected by microsporidia. Five microsporidia inducing xenoma in infected fish off Tunisian coasts were examined for the development of xenomatous structure. Three of these species belong to the genus *Glugea*, one to the genus *Spraguea* and one to the genus *Microgemma*. For each species different, stages of xenoma were examined under light and electronic microscope. For all these species the parasite develops in direct contact with the cytoplasm, which hypertrophy when the number of spores increases. At the periphery the xenoma were first limited by the plasma membrane of the host cell, then, layers of collagen fibres were deposited. After that, fibroblast cells start deposition in a large number of layers.

Immunolocalization of protein kinase A in *Schistosoma mansoni* cercariae and schistosomules (P139)*

Natasha Hirst, Scott Lawton, Anthony J Walker
School of Life Sciences, Kingston University, KT1 2EE, UK

Schistosomes are parasitic blood flukes which are the causative agent of schistosomiasis a disease that affects over 200 million people worldwide, with a mortality rate of over 250,000 per year. This study focuses on protein kinase A (PKA), a signalling enzyme used by eukaryotes to control central cellular functions, to expand current knowledge on the activity of this kinase in schistosomes to benefit the development of potential schistosomicidal drugs. Infective larval stages of *Schistosoma mansoni* were studied. Western blotting of protein lysates of cercariae and 24 hour schistosomules using anti-phospho-PKA (Thr198) antibodies revealed an immunoreactive band at approximately 43 kDa confirming the presence of phosphorylated (activated) PKA in these life-stages. Confocal laser scanning microscopy of cercariae stained with this antibody showed the presence of activated PKA in the tegument, nervous system, and sensory structures at the tip of the cercarial body. Furthermore considerable staining could be seen in specific areas of the cercariae nervous system using anti-phospho PKA substrate antibodies. Schistosomules treated with forskolin, a compound which activates adenylyl cyclase and raises levels of cAMP, displayed increased PKA activation; in addition, schistosomules showed increased rates of contractile movement suggesting that PKA has a role in neuromuscular processes. Our on-going research is investigating the extent to which human dopamine, serotonin and serum affect levels of activated PKA in schistosomules

***Leishmania* Metacyclogenesis Is Promoted in the Absence of Purines (P140)**

Tiago Donatelli Serafim1, Amanda Braga Figueiredo1, Pedro Augusto Carvalho Costa1, Eduardo Almeida Marques-da-Silva1, Ricardo Gonçalves1, Sandra Aparecida Lima de Moura2, Nelder Figueiredo Gontijo3, Sydnei Magno da Silva4, Marilene Suzan Marques Michalick4, Jose´ Roberto Meyer-Fernandes5, Roberto Paes de Carvalho6, Silvia Reni Bortolin Uliana7, Juliana Lopes Rangel Fietto8, Lu´ıs Carlos Crocco Afonso1
1 Laboratório de Imunoparasitologia, 2 Laboratório de Imunopatologia, Departamento de Ciências Biológicas, Núcleo de Pesquisas em Ciências Biológicas, Universidade Federal de Ouro Preto, Ouro Preto, Brazil, 3 Laborato´rio de Fisiologia de Insetos Hematófagos and 4 Laborató rio de Sorologia, Departamento de Parasitologia/ICB, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil, 5 Laboratório de Bioquímica Celular, Instituto de Bioquímica Médica, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil, 6 Laboratório de Neurobiologia Celular, Departamento de Neurobiologia/Programa de Neurociências, Universidade Federal Fluminense, Niterói, Brazil, 7 Departamento de Parasitologia, Instituto de Ciências Biomédicas, Universidade de Sa˜o Paulo, São Paulo, Brazil, 8 Laboratório de Infectologia Molecular Animal, Departamento de Bioquímica e Biologia Molecular/Bioagro, Universidade Federal de Viç osa, Viçosa, Brazil

Metacyclogenesis is important in the life cycle of trypanosomatids as the stage in which infective forms are generated. However, little is known about this fact. This work investigated the effects of adenosine in the *in vitro* and *in vivo* metacyclogenesis of *Leishmania* parasites. It has been generally stated that ''stressful conditions'' will lead to development of metacyclic forms, and with the exception of a few studies no detailed analysis of the molecular nature of the stress factor has been performed. In this work we show that presence/absence of nucleosides, especially adenosine,

controls metacyclogenesis both *in vitro* and *in vivo*. We found that addition of an adenosine-receptor antagonist to *in vitro* cultures of *Leishmania amazonensis* significantly increases metacyclogenesis, an effect that can be reversed by the presence of specific purine nucleosides or nucleobases. Our results show that proliferation and metacyclogenesis are independently regulated and that addition of adenosine to culture medium is sufficient to recover proliferative characteristics for purified metacyclic promastigotes. More importantly, we show that metacyclogenesis was inhibited in sand flies infected with *Leishmania infantum chagasi* that were fed a mixture of sucrose and adenosine. Our results fill a gap in the life cycle of *Leishmania* parasites by demonstrating how metacyclogenesis, a key point in the propagation of the parasite to the mammalian host, can be controlled by the presence of specific purines.

First time reported parasites of equines in Pakistan: Necropsies findings (P141)

Khurram Goraya1, Eugene T. Lyons2
1 Department of Microbiology, University of Health Sciences, Lahore 54600, Pakistan, 2 Department of Veterinary Science, Gluck Equine Research Centre, University of Kentucky, USA

Gasterophilids, habronemids, filariids and cyathostomids are among the most prevalent parasites of equines worldwide. They are frequently responsible not only for ill-thrift, but also for gastro-intestinal dysfunctions including colic and the potentially fatal condition of acute larval cyathostomosis. Six postmortems (two horses, two donkeys and two mules) were performed on euthanized cases. The animals were euthanized in a humane way and the complete gastro-intestinal contents were examined. Parasites were collected and for the identification of species were observed grossly and under the microscope. We found *Gasterophilus nasalus, Habronema muscae, Setaria equina, Cyathostomum catinatum,* and *Cylicocyclus nassatus* were the most abundant parasites among the equines studied. Interestingly, except for cyathostomes, the rest of the reported parasites were insects or insect borne. We conclude that the parasites documented in this study are not new to science but, as far as could be ascertained, are new records from Pakistan. By controlling flies (bot fly, house fly and mosquito), a lot of internal parasites of equines can also be minimized. Euthanized animals were under the treatment of Brooke Hospitals for Animals and had the history of deworming, so anthelmintic resistance is also questionable among reported parasites, especially cyathostomes, there were a lot of them.

Detection of trypanosomes in British badgers (P142)*

Eze Justin Ideozu, Geoff Hide
The University of Salford Manchester School of Environment and Life Sciences Salford M5 4WT

The Eurasian Badger (*Meles meles*) is a popular animal in the UK that is statutorily protected. It has been the subject of intense public health concern resulting from its role as a wildlife reservoir for tuberculosis. Trypanosomes are blood parasites that infect a wide range of hosts, including humans, and have the potential to cause disease in mammals such as badgers perhaps resulting in declining populations. The objectives of this study were to detect trypanosomes in UK badgers using novel molecular biological diagnostic tools. A total of 82 badger blood samples were examined by ITS-PCR using a set of nested primers that targeted the ribosomal RNA gene locus. Twenty-nine of the samples were found to be positive for trypanosomes giving a prevalence of 35.4%. Analysis of sequence data so far confirmed the badgers were infected with *Trypanosoma* (*Herpetosoma*) *otospermophili* and phylogenetic analysis from this study supports the belonging of *T. otospermophili* in the *Herpetosoma* subgenus (NJ and ML tree; 100% bootstrap support). These results show that a significant proportion of UK badgers could be infected with trypanosomes indicating they are susceptible to infection with pathogens. Trypanosome infections in badgers are mediated by transfer of blood by fleas and high prevalence indicates this may be happening frequently. The possibility exists that other important pathogenic diseases, such as tuberculosis, could be transmitted in similar ways. This study could give an insight into the general transmission of infectious diseases in this important wildlife reservoir. Future work is aimed at sequencing other ITS regions.

Implementing the RT-PCR Methodology to Support the Discovery of Gametocytocidal Agents (P143)

Noemi Bahamontes Rosa, María de Gracia Gomez-Lorenzo, Joël Lelièvre, Ane Rodriguez Alejandre, María Jesus Almela, Sonia Lozano, Esperanza Herreros, Francisco Javier Gamo-Benito.
Diseases of the Developing World. GlaxoSmithKline

Although the symptoms of malaria illness is caused by asexual blood stages of the protozoan parasite *Plasmodium* spp., the presence of sexual stages, also called gametocytes, is directly responsible for the infection of the anopheline vector, thus perpetuating the plasmodial cycle. The fight against these transmissible parasite forms is thus an essential strategy

to block transmission that is one of the main goals in the eradication agenda. Currently, there are few methods available for addressing the *in vitro* activity of new antimalarial drugs against gametocytes and persists the need to elucidate if those active compounds have the desired cidal activity. Within this work, we validated a RT-PCR medium throughput technique to successfully identify agents active against mature gametocytes monitoring the level of RNA of genes specifically transcribed at mature sexual stages: Pf77, ROM3, Pfs25 and Pfg377. Those genes were identified by generating a snapshot of the expression profile of 14 candidate genes during a controlled gametocytogenesis. We also intended to discriminate the gametocytocidal activity of standard antimalarial drugs. The clear identification of four sexual mature stage-specific genes that differ from those present in asexual forms and young gametocytes has proven to be a useful tool in antimalarial drug discovery to identify active molecules against mature gametocytes using RT-PCR.

Infection and immunity in the wild house mouse, *Mus musculus domesticus* (P144)

Stephen Abolins1, Louise Hughes1, Laura Weldon1, Liz King2, Julius Hafalla2, Eleanor Riley2 and Mark Viney1
1 School of Biological Sciences, University of Bristol; 2 London School of Hygiene and Tropical Medicine

Mice have been extensively used in laboratory-based immunological and parasitological studies but the immune function and infection status of wild mice is largely unknown. Laboratory mice are usually pathogen free, both for reasons of husbandry but also because pathogens have immunological effects. There has been very little study of the infections and immune function of wild mice. We have investigated common pathogens of some 400 wild mice, specifically ectoparasites, pinworms and several viral and one bacterial infection. Preliminary analysis has found a positive association between total plasma IgG titres and exposure to viral infection, while negative associations are seen between mite infestation and measures of body condition. Infection with the pinworm *Syphacia* spp was common (but absent from mice on Skokholm island and the London Underground). There was a weak, counter-intuitive relationship between intensity of infection and size of these worms; there was no relationship between antibody titres and intensity of infection. Further analyses are now being conducted using structural equation modeling to better understand the intricate interactions between infection and immunity in wild mice.

Sec16 determines the size and functioning of the Golgi in the protist parasite, *Trypanosoma brucei* (P145)

Marco Sealey-Cardona1, Katy Schmidt2, Lars Demmel1, Tatjana Hirschmugl3, Tanja Gesell1, Gang Dong1 and Graham Warren1
1 Max F. Perutz Laboratories, University of Vienna, Medical University of Vienna, Dr. Bohr Gasse 9, 1030 Vienna, Austria, 2 Department of Cell Biology and Ultrastructure Research, Center for Anatomy and Cell Biology, Medical University of Vienna, Schwarzspanierstrasse 17, 1090 Vienna, Austria, 3 Research Center for Molecular Medicine of the Austrian Academy of Sciences (CeMM), 1090 Vienna, Austria.

In the early secretory pathway newly-synthesized proteins and lipid cargo leave the ER in COPII secretory vesicles often from a structure termed the ER exit site (ERES). These cargoes are subsequently processed by the Golgi, mostly by modification of bound oligosaccharides until they reach the trans-Golgi network (TGN), where they are sorted and transported to their final destination. The ERES and Golgi must, therefore, be functionally, and are also often spatially, connected. This relationship must be coordinated so as to fulfill the cell's secretory requirements during the cell cycle. The protist parasite *Trypanosoma brucei* represent a good model system to study this pathway. It has a streamlined architecture, it contains only one ERES and an adjacent Golgi stack at a precise location in the cell between the nucleus and the flagellar pocket. The efficient transport of cell surface coat proteins through the secretory pathway is integral to their success as parasites. How the parasite regulates the trafficking from the ER to the plasma membrane is an important question. Using several approaches we have identified a Sec16 homologue in *T. brucei* – it is a large peripheral protein involved in the formation of COPII vesicles, mediating the transport between the ER and the Golgi. Phenotypic analysis of *T. brucei* following Sec16 depletion or overexpression revealed an intriguing link between secretory capacity and organelle size. Our data suggest that the size of the secretory organelles is optimal for rapid growth of the parasite.

Molecular identification of *Echinostomatidae* of medical and veterinary importance in the UK (P146)*

Egie Elisha Enabulele1,2, Scott P. Lawton1, Anthony J. Walker1, Ruth S. Kirk1
1 School of Life Sciences, Kingston University, Kingston-upon-Thames, UK, 2 Department of Animal and Environmental Biology, University of Benin, Benin City, Nigeria

Molecular DNA sequencing techniques have enhanced the ability to accurately identify members of the

Echinostomatidae which are taxonomically problematic due to poor systematic and phylogenetic resolution of morphological characteristics. The majority of molecular taxonomic studies of echinostomatids have employed a combination of nuclear markers, particularly those associated with ribosomal genes (18S, ITS, 28S), alongside mitochondrial markers including cox1, the international bar-coding gene, and more recently the hyper-variable NAD1. As part of an ongoing molecular study of digenean diversity in lymnaeid snails from freshwater sites in the UK, the echinostomatid species in four lymnaeid snails, *Lymnaea stagnalis, Stagnicola palustris, Radix balthica* and *R. auricularia*, were investigated using the NAD1 genetic marker. Phylogenetic analysis produced well supported clades which identified the echinostomatids to species level. Two medically important species, *Echinostoma revolutum* and *Hypoderaeum conoideum*, and two species of veterinary significance, *Echinoparyphium recurvatum* and *Echinoparyphium aconiatum*, were identified. However, *E. aconiatum* appeared as a sister group to the 37 collar spine *E. revolutum*. *Echinostoma revolutum* is probably widely distributed as it occurred in seven out of nine sites investigated and uses all four snail species as intermediate hosts.

What does control flagellum length during trypanosome development *in vivo*? (P147)*

Eloïse Bertiaux, Sylvie Perrot, Brice Rotureau, Philippe Bastin
Institut Pasteur, France

African trypanosomes are protozoan parasites responsible for sleeping sickness and transmitted by the bite of the tsetse fly. During their complex life cycle, parasites exhibit important morphological variations, especially in flagellum length. Like in other eukaryotes, intraflagellar transport (IFT) is essential for the flagellum construction. IFT is the movement of protein complexes sandwiched between the membrane and the microtubules. From observations in Chlamydomonas, it has been proposed that the amount of IFT material in a given flagellum is proportional to its final length and remains constant at all stages of its elongation. Here, we investigated IFT during *Trypanosoma brucei* development to challenge this model in a situation where the flagellum naturally elongates. Expression and localization of IFT proteins was monitored by immunofluorescence using anti-IFT antibodies during cyclical development of *T. brucei* in *Glossina morsitans*. Remarkably, analysis of fluorescence intensities revealed that the total amount of IFT material in the flagellum is correlated with the length of the flagellum during the natural cycle of the parasite. However, the IFT concentration observed within the flagellum appears more abundant in two specific stages: the short epimastigote and the trypomastigote procyclic forms. Interestingly, these two forms are the only stages where the flagellum will naturally elongate during the developmental process. To confirm these results, trafficking of IFT proteins along flagella will be analyzed in live cells and compared in each stage to propose a model for IFT dynamics related to the natural variations in flagellum length.

The roles of XPC and CSB genes in DNA repair and cell cycle progression in trypanosomes (P148)*

Isabela Mendes1,3, Joao-Pedro V. Da Rocha 1, Simone Calderano 2, Maria C. Elias 2, Carlos R. Machado 1, Richard McCulloch 3
1 Laboratório de Genética Bioquímica, Departamento de Bioquímica e Imunologia, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Brazil 2 Laboratório Especial de Ciclo Celular, Instituto Butantan, Sao Paulo, Brazil 3 Wellcome Trust Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, MVLS, University of Glasgow

XPC and CSB proteins participate in the initial steps of the Nucleotide Excision Repair (NER) pathway, responsible for detecting and repairing lesions that alter DNA conformation. The extent of the genome of trypanosomatids that is transcribed is highly unusual, since most genes are co-transcribed in multigene transcription units, each from a single promoter. The aim of this study was to evaluate the roles of XPC and CSB in *T. cruzi* and *T. brucei* cell cycle and DNA repair. With this purpose, we generated *T. cruzi* single knockouts strains for XPC (XPC+/-) and CSB (CSB+/-), and the same genes were silenced by RNA interference in *T. brucei*. Currently, our data show that *T. cruzi* XPC and CSB single knockouts strains present altered cell cycles and resistance to genotoxic agents. While XPC+/- cells shows delayed cell cycle progression and multinucleated cells, CBS+/- cells show a faster cell cycle and no changes in DNA content. CSB+/- cells were more sensitive to MMS and UV treatments, unlike XPC+/- parasites. RNAi of CSB in *T. brucei* led to increased sensitivity and altered mRNA levels after treatment with UV and cisplatin, while RNAi of XPC was found to be lethal and causes elevated sensitivity to cyclophoshamide. These findings reinforcing the hypothesis that DNA repair genes are involved in cell cycle progression and demonstrate substantial divergence of NER in trypanosmatids, suggesting this may be the best example of any genome repair machinery being a potential drug target against trypanosomatid parasites.

A high-throughput approach for anti-trypanosomal drug target discovery (P149)*

Daniela Begolo, Esteban Erben, Christine Clayton
Zentrum für Molekulare Biologie (ZMBH) Heidelberg University Im Neuenheimer Feld 282 69120 Heidelberg, Germany

High-throughput screens are revealing novel chemical classes with anti-kinetoplastid activity. To improve the efficacy of drug candidates it is useful to know the drug target, since knowledge of the way a drug interacts with its target can be used to guide the medicinal chemistry and can assist in product registration. We established a large inducible overexpression library for trypanosomes, containing random genomic DNA fragments. The library was transfected into *Trypanosoma brucei* bloodstream form. A library of transformed parasites was obtained, large enough to ensure several-fold in-frame coverage of protein-coding regions. The overexpression library was used in cultures containing drugs. The surviving trypanosomes were screened to map which proteins were capable of conferring growth advantage. The resulting cloned fragments could encode direct targets of the drugs, or at least aid in clarifying the drug's mode of action. As a proof of principle, the approach was validated with two drugs whose targets are already known, difluoromethylornithine and DDD85646. The library will be used with drugs under development to look for new possible drug targets.

Synergistic trypanocidal effect of the cysteine protease inhibitor K11777 and eflornithine (P150)

Dietmar Steverding
BioMedical Research Centre Norwich Medical School University of East Anglia Norwich NR4 7TJ

Chemotherapy of human African trypanosomiasis (HAT) is unsatisfactory because only a few drugs, with serious side effects and poor efficacy, are available. Because drug combination regimes often achieve greater therapeutic efficacy than monotherapies, we investigated the trypanocidal activity of the cysteine protease inhibitor (CPI) K11777 in combination with current anti-HAT drugs using bloodstream forms of *Trypanosoma brucei.* Whereas combinations of K11777 with either suramin, pentamidine or melarsoprol showed antagonistic effects with a combination index (CI) of 1.67, 1.46 and 1.46, respectively, the combination of K11777 with eflornithine (DFMO) showed a synergistic effect with a CI of 0.67. As eflornithine inhibits the polyamine biosynthesis leading to a decrease in trypanothione levels, and as the activity of the targeted lysosomal cysteine protease (TbCATL) is dependent on thiols, it is reasonable to suggest that the synergistic effect of the K11777/DFMO combination is associated with a reduced thiol concentration in the lysosome. To prove this, we tested another CPI, CA074Me, which inhibits TbCATL only under reducing conditions, in combination with eflornithine. The CA074Me/DFMO combination showed an antagonistic effect with a CI of 1.33. In contrast, CAA0225, a CPI related to CA074Me, which inhibits TbCATL independently of thiols, showed a synergistic effect in combination with DFMO with a CI of 0.89. These results confirm the suggestion that the underlying mechanism for synergy of the K11777/DFMO combination is indeed facilitated by the reduction in thiol levels. Encouragingly, the K11777/DFMO combination that inhibited the growth of trypanosomes by 50% had no cytotoxic effect on human HL-60 cells.

Developing Peptide-Mimetics to Treat Cutaneous Leishmaniasis (P151)

Gabriela Eggimann, Hannah L. Bolt, Alexandra M. Webster, Paul W. Denny, Steven L. Cobb
Department of Chemistry, Durham University, UK; School of Biological and Biomedical Sciences, Durham University, UK

Antimicrobial peptides (AMPs) have been proposed as one potential solution to the development of new topical anti-leishmanials to treat cutaneous Leishmaniasis (CL). However, their inherent chemical and biological instability presents a major hurdle and only a few AMPs are currently in clinical trials for the treatment of skin infections. Early progress with synthetic peptide mimetics suggests that these molecules offer a significantly better opportunity for development from bench to market. Amongst the AMP mimetics reported, peptoids have considerable potential for the development of new topical anti-infective agents. Relative to AMPs, antimicrobial peptoids are cheaper to manufacture and have significant therapeutic potential as a consequence of their structural stability, superior bioactivity and resistance to protease degradation. They also retain broad spectrum activity against multidrug resistant bacterial strains, as shown in literature. We have recently identified for the first time selected peptoids with anti-parasitic activity against *Leishmania mexicana* promastigotes and amastigotes causing CL (unpublished). These peptoids are active in the low µM range and were used as leading compounds in a structure-activity-relationship (SAR) study to elucidate the mode of action against the parasite and their toxicity against skin cells. Here we will present the work we have carried out to develop peptoids as an entirely new and promising class of anti-leishmanial agents.

Genomic ancestry blocks decipher population gene flow and admixture in monomorphic *Leishmania donovani* (P152)
Downing T1, Imamura H2,3, Mannaert A2, Vanaerschot M2, Stark O4, Schonian G4, Dujardin JC2, Berriman M3, Cotton JA3.
1 *College of Science, National University of Ireland, Galway, Ireland; 2 Institute of Tropical Medicine, Antwerp, Belgium; 3 Wellcome Trust Sanger Institute, Cambridge, UK; 4 Charitè University, Berlin, Germany.*

Mixing between genetically distinct pathogen strains leads to novel offspring with altered host virulence and drug susceptibility. Newly isolated genomes may represent undiscovered lineages or re-assortments between known groups. Genomic and statistical investigation of variation discovered in new genomes compared to reference genotypes provides a framework for determining their ancestry and predicting phenotypes. Current methods of allele frequency correlation, variant distribution modality and admixture modelling are effective for contexts where subspecies interbreed, but are as yet untested for monomorphic populations where informative mutations are rare. Haplotype distribution, size and length decay provides sufficient power to distinguish diploid sample pairs with a mean of just 3.4 pairwise SNPs/Mb in a sample of 191 clinical isolates of Indian subcontinent *Leishmania donovani* sampled in 2002-11 during two drug treatment eras. Population clustering models identified six major genetically homogeneous populations within Nepal and India with little evidence of recent interbreeding: their unique ancestral haplotypes were profiled. Population-free membership assignment, phylogenetic and admixture D/F-statistics indicated six recent Indian isolates were discovered whose haplotypes were mixes of distinct populations. This general population genomic approach can distinguish populations from mixtures with just 60 genome-wide SNPs between diploid parasite groups. Notably, seven samples had a haplotype structure did not resemble any previous sample: these represented either rare/undersampled lineages only identifiable from haplotype-based rather than SNP-based investigation. These insights into recent gene flow identify the tendency for different populations causing leishmaniasis to mate, in which the potential for hybrid vigour to cause new outbreaks is currently unknown.

Aquaglyceroporin 2 mutations in *Trypanosoma brucei gambiense* field isolates are linked to drug resistance (P153)*
Fabrice E. Graf1,2, Philipp Ludin1,2, Tanja Wenzler1,2, Marcel Kaiser1,2, Reto Brun1,2, Patient Pati Pyana3,4, Philippe Büscher4, Harry P. de Koning5, David Horn6, Pascal Mäser1,2
1 Swiss Tropical and Public Health Institute, Basel, Switzerland; 2 University of Basel, Basel, Switzerland; 3 Institut National de Recherche Biomédicale, Kinshasa Gombe, Democratic Republic of the Congo; 4 Institute of Tropical Medicine, Antwerp, Belgium; 5 University of Glasgow, Glasgow, United Kingdom; 6 University of Dundee, Dundee, United Kingdom

The predominant mechanism of drug resistance in *Trypanosoma brucei* is decreased drug uptake due to loss-of-function mutations in transporters that mediate drug import. One such transporter is the well-characterized aminopurine permease P2, encoded by TbAT1, which transports melarsoprol, pentamidine, and diminazene. TbAQP2 has just recently been identified to play a major role in the well-known melarsoprol/pentamidine cross-resistance. The role of transporters as determinants of drug susceptibility is well documented from laboratory-selected *Trypanosoma brucei* mutants but not from clinical isolates. 16 *T. brucei* ssp. field isolates that (i) have been adapted to axenic *in vitro* cultivation and (ii) in several cases stem from melarsoprol treatment-refractory cases have been genotyped regarding TbAT1 and TbAQP2 and the drug sensitivities have been determined for melarsoprol, pentamidine and diminazene. We found mutations in the TbAQP2 / TbAQP3 locus in several *T. b. gambiense* field isolates, leading to the formation of a novel type of TbAQP2-TbAQP3 chimera. The identified mutant *T. b. gambiense* are all 40- to 50-fold less sensitive to pentamidine and 3- to 5-times less sensitive to melarsoprol than reference isolates. We show for the first time that mutations in the TbAQP2/TbAQP3 locus accompanied by TbAQP2 gene loss also occur in the field, and that the *T. b. gambiense* carrying such mutations correlate with a significantly reduced susceptibility to pentamidine and melarsoprol and might be responsible for melarsoprol treatment failures. Currently, we are investigating the function of the TbAQP2-TbAQP3 chimera with respect to drug transport in a TbAQP2 null line.

Probing druggability and biological function of essential proteins in *Leishmania* combining facilitated null mutant and plasmid shuffle analyses (P154)
Mariko Dacher1, Miguel A. Morales1, Pascale Pescher1, Olivier Leclercq1, Eric Prina1, Mathieu Cayla1, Albert Descoteaux2, Gerald F. Späth1
1 Institut Pasteur, CNRS URA 2581, Unité de Parasitologie moléculaire et Signalisation, Paris, France; 2 INRS-Institut Armand-Frappier and Center for Host-Parasite Interactions, Laval, Québec, Canada

Leishmania parasites cause important human morbidity and mortality. Essential *Leishmania* genes escape genetic assessment by loss-of-function analyses due to lethal null mutant phenotypes, even though these genes and their products are biologically most significant and represent validated drug targets. Here we overcome this limitation using a facilitated null mutant approach applied for the functional genetic analysis of the MAP kinase LmaMPK4. This system relies on the episomal expression of the target gene from vector pXNG that expresses the Herpes simplex virus thymidine kinase gene thus rendering transgenic parasites susceptible for negative selection using the antiviral drug ganciclovir. Using this system we establish the genetic proof of LmaMPK4 as essential kinase in promastigotes. LmaMPK4 structure/function analysis by plasmid shuffle allowed us to probe for LmaMPK4 druggability and to establish a partial null mutant expressing an MPK4 derivative with altered ATP binding properties. This mutant showed an effect in metacyclogenesis, linking for the first time MPK4 function with a crucial parasitic function. The facilitated knock out approach allowed us to validate MPK4 as an essential protein kinase, to identify regulatory kinase sequence elements relevant for chemotherapeutic intervention and to link MPK4 functions to metacyclic differentiation. The approaches presented here are broadly applicable to any essential gene in *Leishmania* thus overcoming major bottlenecks for their functional genetic analysis and their exploitation for structure-informed drug development.

Identification of genes essential for viability in *Leishmania* amastigotes - N-myristoyltransferase as a model (P155)

Daniel Paape1,2, Catriona T. Prendergast1, Helen P. Price1, Johannes Doehl1, Deborah F. Smith 1
1 Centre for Immunology and Infection, Department of Biology/Hull York Medical School, University of York, YO10 5DD, UK; 2 current address: Wellcome Trust Centre for Molecular Parasitology; Institute of Infection, Immunity and Inflammation; University of Glasgow; 120 University Place, G12 8TA, UK

Proving that specific genes are essential for intracellular viability in *Leishmania* remains a challenge, especially in the absence of a robust inducible expression system or a functioning RNAi machinery that works in all *Leishmania* spp. At present, if a target gene of interest can only be deleted from its genomic locus in the presence of ectopic expression in extracellular promastigotes, it is assumed to be essential for viability, with these data often correlated with similar function in intracellular amastigotes. Ideally, functional essentiality must be proven independently in both life cycle stages, particularly if the gene under study is a putative drug target. N-myristoyltransferase (NMT) has been shown to be essential in promastigotes of *Leishmania major* and *L. donovani* and is being exploited for the development of chemotherapeutic intervention in human leishmaniasis. However, to date, it has not been shown that NMT is essential for the viability of intracellular amastigotes. Plasmid shuffle technology, first described in *Saccharomyces* spp., is based on the concept that a target gene-carrying plasmid can only be lost if another functional copy of the gene or necessary nutrients are present. We have used plasmid shuffle, previously used in promastigotes only, to provide *in vivo* proof for the first time that the NMT protein is essential in both promastigotes and amastigotes, hence confirming its validity as a target for drug development in *Leishmania*.

Homologous transcript interference maintains VSG allelic exclusion in African trypanosomes (P156)*

Sebastian Hutchinson1, Sam Alsford2, Lucy Glover1, David Horn1
1 *University of Dundee; 2 London School of Hygiene and Tropical Medicine*

African trypanosomes display monoallelic transcription of a telomeric variant surface glycoprotein (VSG) gene. Other VSGs are reversibly silenced and VSG switching elicits antigenic variation in approximately 0.001% of cells per cell division. We have found that VSG allelic exclusion operates through potent interference among homologous sequences. We first show that artificial transcription blockade of the single active VSG triggers activation of silenced VSGs in more than 10% of cells, representing a 10,000 fold increase in switching frequency. This negative feedback control impacts VSGs inserted at both telomere-adjacent loci and non-telomeric loci and strength of repression correlates with sequence-similarity in the region representing the 3' untranslated region (3'-UTR) of the active VSG transcript. Interference can also be mediated by other homologous sequences, as demonstrated by a synthetic negative feedback circuit comprising exogenous reporters with common 3'-UTRs. Finally, knockdown of the active VSG RNA triggers activation of silent VSGs. We conclude that VSG transcripts, and likely other VSG-associated transcripts, mediate sequence-specific allelic interference, thereby maintaining allelic exclusion.

Cholinergic signaling and the respiratory immune response to parasitic nematode infection (P157)*

Luke Roberts, Corinna Schnoeller, Murray E. Selkirk
South Kensington Campus, Imperial College London, UK

A number of nematode parasites secrete acetylcholinesterase enzymes (sAChE) within the host. These enzymes break down acetylcholine (ACh), hypothesised to play an important role as a co-stimulator or mediator of immune responses. With this in mind, we sought to characterise cells which are involved in immunity to nematode infection and capable of producing ACh, in order to ascertain whether these cells are the targets of helminth derived sAChE. Using the *Nippostrongylus brasiliensis* model of nematode infection, and an eGFP-reporter mouse strain which identifies cells capable of synthesising ACh, we were able to demonstrate that a number of hematopoietic cells in the lung significantly up-regulate their capacity for ACh synthesis following infection, with some interesting differences seen both in the early, acute stage of inflammation and later chronic stage of tissue repair. Among these, type-2 innate lymphoid cells (ILC2) and effector/memory CD4+ Th2 cells displayed the most striking increases. Mass spectrometry was used to quantify ACh release from sorted respiratory cell populations while RT-PCR was used to further characterise expression of cholinergic receptors and other features of ACh producers. Given the emerging role of ILC2 in allergy and respiratory disease such as asthma and COPD (which display a similar immune-pathology to anti-helminth immunity) and the current use of anti-cholinergic drugs to treat a number of these pathologies, exploring the production of ACh by immune cells and its downstream signalling effects may not only reveal new information about host-parasite interactions, but may provide new targets for treatment of respiratory illnesses.

Endotoxin in human *T.b.rhodesiense* infection (P158)

Jeremy Sternberg, Eltayb Abdelwahab Aboubaker
Institute of Biological and Environmental Science, University of Aberdeen, Zoology Building, Aberdeen AB24 2TZ

Elevated endotoxin levels have previously been reported in the plasma and CSF of *T. b. gambiense* sleeping sickness patients and also in the serum of *T. b. brucei*-infected experimental mice. There have been conflicting data on the source of the endotoxin, and in particular as to whether it is LPS from gram negative bacteria resulting from histopathological lesions to the gut during infection or wether it derives from an endotoxin-like substance occurring in the trypanosome. We have used a set of human *T. b. rhodesiense* infection plasma and CSF with associated detailed clinical pathology data to address this question. Endotoxin activity was significantly increased in plasma during infection, and this declined after chemotherapy. There was no relationship between parastiaemia and endotoxin level. Furthermore, no increase in endotoxin levels in the CSF were detected following CSN invasion by the parasites. Our results provide indirect evidence that the endotoxin in this infection is derived from microbial sources, probably in the gut. Entotoxin is potentially an important pathophysiological factor in infection, and there was a significant relationship between the levels of endotoxin and those of the inflammatory cytokine IL-6.

Cell Surface Proteome Analysis of *Trypanosoma cruzi* Life Stages (P159)

Sébastien Charneau1, Rayner Myr Lauterjung Queiroz1, Flávia Nader Motta2,3, Izabela Marques Dourado Bastos3, Jaime Martins de Santana3, Marcelo Valle de Sousa1, Peter Roepstorff4, Carlos André Ornelas Ricart1.
1 Laboratory of Biochemistry and Protein Chemistry, Department of Cell Biology, Institute of Biology, University of Brasilia, Brasília, 70910-900, Brazil, 2 Faculty of Ceilândia, University of Brasilia, Brasilia, 70910-900, Brazi, 3 Laboratory of Host-Pathogen Interactions, Department of Cell Biology, Institute of Biology, University of Brasilia, Brasília, 70910-900, Brazil, 4 Department of Biochemistry and Molecular Biology, University of Southern Denmark, Campusvej 55, Odense, DK-5230, Denmark.

Chagas disease is a neglected infectious illness, caused by the protozoan *Trypanosoma cruzi*. It remains a challenging health issue since there is no immunoprophylatic vaccine or satisfactory chemotherapy treatment for its chronic stage. The present work addressed the analysis of *T. cruzi* plasma membrane (PM) subproteome from epimastigote insect life stage and trypomastigote and amastigote human-hosted life stages. Two complementary PM protein enrichment techniques followed by identification using a nanoLC-MS/MS (LTQ-Orbitrap Velos) approach were optimized and evaluated from epimastigotes and then applied to tissue culture-derived trypomastigotes and axenic amastigotes. The first methodology was based on cell surface trypsinization (Shave) of intact living cells while the second approach used biotinylation of cell surface proteins followed by streptavidin affinity chromatography isolation of the labeled proteins. The results revealed an extensive repertoire of proteins in the PM subproteomes, including enzymes that might be suitable candidates for drug intervention. The comparison of the cell surface proteome among the human-hosted life forms revealed some potentially stage-specific enzymes, although the majority was shared by both stages. Bioinformatic analysis showed that the vast majority of the identified proteins are membrane-derived and/or possess predicted transmembrane domains. They are mainly involved in host cell infection, protein adhesion, cell signalling and the

modulation of mammalian host immune response. Several virulence factors and proteins potentially capable of acting at a number of metabolic pathways of the host and also to regulate cell differentiation of the parasite itself were also found.

An RNAi screen for drug resistance in African trypanosomes reveals a link between acidic compartment function and mitochondrial F0F1 ATPase function (P160)

Nicola Baker, Michael P Barrett2, Jonathan Wilkes2, Graham Hamilton2 and David Horn1
1 Division of Biological Chemistry & Drug Discovery, College of Life Sciences, University of Dundee, Dow Street, Dundee DD1 5EH 2 Wellcome Trust Centre of Molecular Parasitology, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow G12 8TA

Isometamidium (ISM) is one of the most important veterinary drugs currently used for the treatment of African trypanosomiasis or nagana in livestock, but drug resistance is an increasing problem. This drug accumulates in the trypanosome kinetoplast which encodes a single essential F_0F_1-ATPase subunit in bloodstream-form *T. brucei*. We used genome-scale RNAi library screening to identify ISM resistance mechanisms. Among twenty-four genes linked to ISM resistance were thirteen V-type ATPase subunits and all four adaptin 3 (AP-3) subunits, strongly implicating acidic compartment (lysosome and acidocalcisome) defects in resistance. Knockdown of these subunits and chemical inhibition of the V-ATPase increased resistance to ISM. Surprisingly, ISM uptake was undiminished following knockdown, but we found that kinetoplast loss, although normally lethal, was well-tolerated. We also found that these cells no longer required the mitochondrial F_0F_1-ATPase and displayed cross-resistance to other kinetoplast-binding drugs. Our results reveal an unexpected link between acidic compartment function and mitochondrial F_0F_1-ATPase function, possibly reflecting cross-talk among the major two-sector ATPases. They also show that V-ATPase or AP-3 mutations could contribute to drug resistance.

Wild rodents and the ectoparasite fauna from coastal and island habitats in Peninsular Malaysia and its public health importance (P161)

Mohd Zain, S.N1, Nur Syazana, M.T1, Behnke, J.M2
1 Institute of Biological Sciences, Faculty of Sciences, University Malaya, 50603 Kuala Lumpur, Malaysia, 2 School of Biology, University of Nottingham, University Park, Nottingham NG7 2RD, United Kingdom.

A study to determine the diversity and distribution of ectoparasites infesting wild rodent populations was carried out between July 2010 and December 2011 in coastal and island habitats in Peninsular Malaysia. A total of 363 rodents were captured with four rodent species identified namely, *Rattus tiomanicus, Rattus rattus diardii, Rattus argentiventer* and *Rattus norvegicus*. Low diversity of ectoparasites was observed infesting the rodent populations with 9 ectoparasite species recorded. The ectoparasites recovered fell under 4 broad taxa, namely; flea (*Xenopsylla cheopis*) mites (*Laelaps nuttali, Laelaps echidninus, Laelaps sculpturatus, Listrophoroides* sp. and *Ornithonyssus bacoti*), lice (*Polyplax spinulosa* and *Hoplopleura pacifica*) and ticks (*Ixodes granulatus*) The general index of *X. cheopis* was low and range between 0.01-0.12. Four of the 9 species recorded have been incriminated as important vectors or mechanical carriers for transmission of zoonotic diseases namely *Ornithonyssus bacoti*; *Hoplopleura pacifica, Polyplax spinulosa* and *Xenopyslla cheopis*. Intrinsic (host age and sex) and extrinsic (location and season) were analysed to understand the factors that influence the prevalence and abundance of ectoparasites on these host populations.

Trypanosoma* specific subunit ATPaseAF2 helps to anchor F_1-ATPase moiety to the mitochondrial membrane(P162)

K. Šubrtová1,2, B. Panicucci2, A. Zíková2
1 Faculty of Science, University of South Bohemia, Branišovská 31, 370 05 České Budějovice, Czech Republic; 2 Laboratory of Functional Genomics of Protists, Department of Molecular Parasitology, Institute of Parasitology, Biology Centre, ASCR, v.v.i. , Branišovská 31, 370 05 České Budějovice, Czech Republic

In the infectious stage of *Trypanosoma brucei*, an important parasite of humans and livestock, the mitochondrial membrane potential($\Delta\psi m$) is uniquely maintained by the hydrolytic activity of the essential F_0F_1-ATPase, which contains several trypanosome specific subunits of unknown function. Here, we determined that one of the largest novel subunits, ATPaseAF2, is membrane-bound and incorporates into monomeric and multimeric assemblies of the F_0F_1-ATPase. RNAi silencing of ATPaseAF2 led to a significant decrease of the $\Delta\psi m$ and consequently to *T. brucei* growth inhibition. To further explore the function of this protein, we employed a naturally occuring trypanosoma strain that lacks

mitochondrial DNA (dyskinetoplastic, Dk) and thus subunit a, an essential component of the Fo-moiety. These Dk cells maintain the Δψm by the electrogenic exchange of ATP4-/ADP3- by the ATP/ADP carrier and the hydrolytic activity of F_1-ATPase. Interestingly, the glycerol gradient sedimentation and native electrophoresis of Dk mitochondria revealed, in addition to F_1-ATPase, the presence of monomeric and multimeric F_0F_1-ATPase. Furthermore, the membrane bound ATPaseAF2 subunit is expressed in Dk cells and co-sediments with these higher sedimenting complexes. RNAi studies demonstrate that the ATPaseAF2 subunit is essential for Dk cells, crucial for maintaining the Δψm and important for the stability of the membrane associated Dk F_0F_1-ATPase. Importantly, electron micrographs revealed that the Dk F_1-ATPase is de-attached from the membrane upon silencing of ATPaseAF2. In conclusion, we propose that ATPaseAF2 is responsible for connecting the Dk F_1-ATPase to the mitochondrial membrane, thus increasing the efficiency of the functional association between F_1-ATPase and ATP/ADP carrier.

Infective stage plant parasitic nematode population density influences the dispersal behaviour of conspecifics (P163)*

Emily Robb1, Ivan Vokřál2, Johnathan Dalzell1, Colin Fleming3, Angela Mousley1, Aaron G. Maule1 and Nikki J. Marks1
1 Institute for Global Food Security, School of Biological Sciences, Queen's University Belfast, UK, 2 Department of Pharmacology and Toxicology, Charles University, Prague, Czech Republic; 3 AgriFood Biosciences Institute, Newforge Lane, Belfast, UK

Plant parasitic nematodes (PPNs) significantly reduce crop production systems such that their control is a significant issue for global food security. Whilst much effort has focused on understanding the interaction between parasitic-stage PPNs and their plant hosts, relatively little effort has centred on understanding the behaviour of pre-parasitic J2 stage PPNs in the soil. Recent data on the chemical ecology of *C. elegans* illustrate the rich capacity for nematodes to influence the behaviour of conspecifics. We aim to develop a better understanding of the behaviour of pre-parasitic J2s to help seed the development of novel control strategies that disrupt soil-based orientation-behaviours. We hypothesise that inter-J2 signalling modulates the behaviour of conspecifics and set out to investigate the influence of population density on dispersal behaviour. The dispersal of worms derived from low density (50 J2s) and high density (500 J2s) populations of *Globodera pallida* or *Meloidogyne incognita* were compared. Worms derived from higher density populations dispersed significantly faster and further than those worms derived from less dense populations, e.g. after 6 hours in pluronic gel, 43% of *G. pallida* J2s derived from high density populations had dispersed compared to only 14% from low density populations; similar results were seen for *M. incognita* J2s. Additional efforts seek to dissect inter-cellular signalling molecules that influence J2 behaviours. Further we have initiated RNA interference experiments that selectively silence PPN sensory signalling components. Initial efforts are focussed on the influence of PPN flp-21 signalling (impacts sociality in *C. elegans*) on worm movement/dispersal.

The impact of anthelmintic treatment on the incidence of diarrheal disease in Vietnamese school children (P164)*

Jacqueline M. Leung1, Phan Vu Tra My2, Stephen Baker2, Andrea L. Graham1
1 Princeton University, Department of Ecology and Evolutionary Biology, Princeton, NJ, USA, 2 Oxford University Clinical Research Unit, Enteric Infections Department, Ho Chi Minh City, Vietnam

Cheap and effective drugs called 'anthelmintics' are currently available to clear parasitic helminths from the human intestinal tract. As a result, mass school-based 'deworming' programs are becoming routine in developing countries and are receiving wide support from international health organizations. However, experimental evidence suggests that anthelmintic treatments may also increase host susceptibility to other infections (bacteria, viruses, or protozoa). This is because anthelmintic treatment reverses the immunomodulatory effects of helminths and opens up ecological niches in the gut microbiota for pathogens to invade. Current deworming programs focus solely on reducing the symptoms of helminth infections and often fail to monitor for unintended consequences of treatment on other pathogens. In fact, most research in biomedicine focuses on single-species infections, despite the ubiquity of co-infections in nature. We have unique preliminary evidence from Vietnam suggesting that helminths may be beneficial to children in combating against diarrheal infections. Thus, we propose to conduct a placebo-controlled trial to examine the effects of anthelmintic treatment on the health of school children and their subsequent risks of diarrheal disease due to viral and bacterial infections. Our results will provide new insights into the role of co-infections in the success of treatment strategies and may thus reshape the design and implementation of deworming programs today.

Effect of season and multiplicity on gametocytes among chronic asymptomatic *Plasmodium falciparum* carriers in absence of transmission (P165)

Amal Gadalla1,2, Petra Schneider3, Elkhansaa Nassir2, Abdel-Muhsin A Abdel-Muhsin2, Thomas Churcher4, Sarah Reece3, Hamza Babiker1
1 College of Medicine, Sultan Qaboos University, Alkhoud 123, Muscat, Oman, 2 Tropical Medicine Research Institute, National Center for Research, Khartoum, Sudan, 3 School of Biological Sciences, University of Edinburgh, EH9 3JT, UK; 4 Department of Infectious Disease Epidemiology, Imperial College London, London, UK

Asymptotic malaria represents a major challenge to control efforts and the prospect of malaria elimination. In areas of marked seasonal malaria, asymptomatic parasitaemia that persists during the transmission-free period can produce gametocytes, and serves as reservoir to raise new cases following annual rains and resurgence of *Anopheles* mosquitoes. The explosive nature of transmission in these areas, suggest that asymptomatic parasitaemia adjust gametocytes production to coincide with appearance of mosquitoes. The current study examined the above hypothesis, and assesses other factors that modulate gametcytogeneis, among asymptomatic carriers, in absence of transmission. We closely monitored parsitaemia, gametocytaemia, using qPCR and multiplicity of infection (pfg377 and msp2) among 38 individuals who sustained asymptomatic *P. falciparum* infection, throughout the dry season in an area of seasonal transmission in eastern Sudan. Monthly data from the above cohort were analysed using GLMM model. Asymptomatic *P. falciparum* parasitaemia that persisted in the dry season, were characterized by (a) low parasite density ranging between 0.014- 4982.8 parasites/µl. (b) high prevalence of multiplicity (64-88%) and (c) gametocytes carriage (7-30%). Interestingly, gametocyte density peaked following the start of rains, coinciding with resurgence of mosquito, when acquisition of new infection can be excluded. Gametocyte density influenced by the interaction between parasite density and multiplicity of infection. Higher gametocyte density was associated with low parasite density when infection contains multiple genotypes.

Further studies on addressing the need of praziquantel in young children (P166)

J.R. Stothard, D. Waterhouse, A. Bustinduy, S. Ward
Liverpool School of Tropical Medicine

Prazqiuantel is an orally administered anthelminthic and used in preventive chemotherapy campaigns for control of schistosomiasis. Praziquantel (Biltricide®) is a racemate drug containing equal amounts of the biological active enantiomer R-praziquantel and the inactive distomer S-praziquantel. The drug is licensed for use in children over the age of four and is typically given out as a single treatment at 40mg/kg bodyweight. Until recently, however, the treatment needs of children younger than four years of age have been overlooked and in certain areas they are living in need of regular treatment. Ongoing studies in Uganda, however, have shown that the parasitological cure and egg reduction rate in infants and preschool-aged children with intestinal schistosomiasis can be poor. To improve antiparasitic performance, different dosing regimes may be needed, for example increasing the dosage to 60mg/kg. To shed light on the dynamics of praziquantel, a pilot study was undertaken in 60 Ugandan children with egg-patent infections who were then treated with either 40 or 60mg/kg. To measure bloodstream levels of R-S-praziquantel, venous blood was collected at 7 time points during a 24hr period. The samples were then processed and analysed by LCMS using chiral separation, owing to the isomeric nature of praziquantel. The quantification of both enantiomers via chiral-LCMS enabled a comprehensive collection of pharmacokinetic parameters for both R/S-enantiomers. In this poster we present information on the assays used in the field and laboratory protocols developed illustrated with preliminary results.

Characterization of trypanosome infections over the lifetime of cattle in Ghana (P167)

Theresa Manful1, Jennifer Afua Ofori1 and Mark Carrington2
1 Department of Biochemistry, Cell and Molecular Biology University of Ghana P. O. Box LG54 legon; 2 Department of Biochemistry Tennis Court Road Cambridge CB2 1QW

Animal trypanosomiasis, caused by different species of trypanosomes affects both domestic and wild animals in sub-Saharan African. The disease has a direct effect on food production and thus a negative impact on economic development in the affected areas. The most common trypanosome species detected in cattle in Ghana are *T. brucei, T. congolense* and *T. vivax* and *T. simiae*, and the prevalence of animal trypanosomiasis ranges from 5-50%. There have not been any studies on lifetime infections with trypanosomes in cattle. We will investigate trypanosome diversity and characterize trypanosomes infections in cattle over time. Two heads of cattle have been selected based on their geographical location, tsetse fly density, prevalence of trypanosomiasis and the breed of cattle available. Blood samples will be taken from the animals at monthly intervals, DNA extracted, and trypanosome species and isolates will be characterized. The data generated from this study will provide invaluable information on the biology of trypanosome

infection and help inform control measures in the areas.

The effect of *Leishmania major* infection on atherogenesis and cytokines pattern in resistant and susceptible mice (P168)

Marc Karam, Mirna Chahine, Amani Chahine
Faculty of Sciences, Biology Department, University of Balamand - Lebanon

The outcome of the infection *Leishmania major* depends on the type of the immune response mounted by the host. This response is either cell mediated or humoral making the host resistant (as humans and C57BL/6 mice) or susceptible (BALB/c mice) to this parasite respectively. Activation of either branch of the immune system depends on other factors such as genetic makeup of the host, the infecting dose of parasites and the cytokine milieu (during early stages of the infection). In BALB/c mice, the early production of Th2 cytokines (TGF-β, IL-4, IL-5, IL-10 and IL-13) inhibits IL-12 function and natural killer (NK) cells which are crucial for Th1 cell activation. On the other hand, *L. major* infection in resistant hosts results in Th1 cells activation with production of IL-2, Interferon-γ (IFN-γ) and Tumor Necrosis Factor-α (TNF-α). On the other hand, inflammation plays an important role in atherogenesis whereby T helper cells seem to play a role in the plaque development. Th1 cells produce mainly IFN-γ, TNF-α and IL-2 which lead to the activation of macrophages, induction of endothelial dysfunction and promotion of plaque destabilization. While Th2 cells produce mainly IL-4, IL-5, IL-13 and IL-25 and seem to have an atheroprotective role because they suppress the Th1 pro-inflammatory response. We hypothesize here that Th1 and proinflammatory cytokines that confer protection against this parasite can promote atherogenesis in resistant hosts while Th2 cytokines might have protective effects against atherosclerosis while making the hosts susceptible to *L. major* infection.

Eco-immunology: thermal variation and immunity in the three-spined stickleback (P169)

Alex Stewart1, Joe Jackson2, Martha Brown2, Chris Williams3, Jo Cable1
1 School of Biosciences, Cardiff University, Cardiff, CF10 3AX, United Kingdom; 2 IBERS, Aberystwyth University, Aberystwyth, SY23 3DA, United Kingdom; 3 Environment Agency, Brampton, Cambridgeshire, PE28 4NE, United Kingdom

Temperatures still within the physiological ranges of ectotherms have been shown to alter immune function. The activity of the adaptive response appears to be highly dependent on ambient temperature with temperate species of fish experiencing a complete loss of function below 10°C. In periods of cold weather, this phenomenon can result in an immunocompromised host, potentially causing death; termed 'winter kill syndrome'. Advances in molecular and genomic techniques coupled with the widespread availability of the three-spined stickleback (*Gasterosteus aculeatus*) have made it an ideal system in which to study eco-immunology in a temperate, but changing, climate. We have used the stickleback genome, published on Ensembl in 2006, coupled with degenerate PCR and de novo sequencing to develop a myriad of Q-PCR primers to target immune markers such as Gata-3, IFN-γ and MHCII. Using this Q-PCR toolbox we can conduct experiments to understand how thermal variation affects stickleback immunology in the context of infection. Our current study exposes sticklebacks to cold (6°C) conditions for different periods before simulating a spring warming to 15°C in order to understand how long it takes the immune system to recover from a non-permissive state. Our aim is to inform fish stock managers on the potential problems associated with climate change, parasitism and immunity and how best to manage infection in a warming climate.

Dynamics of Anti-Glycan Antibody Responses in *Schistosoma japonicum*-Infected Rhesus Macaques Studied by Schistosome Glycan Microarray (P170)*

Y.Y. Michelle Yang1, Angela van Diepen1, Xiao Hong Li2, R. Alan Wilson3, Cornelis H. Hokke1
1 Department of Parasitology, Leiden University Medical Center, Leiden, The Netherlands, 2 Key Laboratory of Parasitology and Vector Biology, MOH National Institute of Parasitic Diseases, Chinese Center for Disease Control and Prevention, Shanghai, China,1 3 Centre for Immunology & Infection, Department of Biology, University of York, York, United Kingdom

Human immunity to schistosome infection requires many years of exposure to the parasite, multiple infections and treatments to develop. Unlike humans, the rhesus macaque is able to rid itself of an established infection, as seen by the deterioration of adult worm accompanied by the decline of egg excretion. The worms are killed as a result of antibody-mediated processes towards undefined molecular targets. In human and animal *S. mansoni* infection, serum antibodies directed against schistosome glycan antigens are induced. To be able to conduct studies on the specificity of these

antibodies and identify the structure of their glycan targets, we have previously created a schistosome microarray containing a large repertoire of N-, O- and lipid glycans isolated from *S. mansoni* larvae, adults worms and eggs. Using this glycan microarray approach, we now analyzed serum IgG and IgM of *S. japonicum*-infected macaques in a longitudinal study of 22 weeks. We found that in the beginning of the infection, antibody response towards schistosome glycans are dominated by IgM. As the infection continues, a profound increase in IgG intensity is observed mainly towards a multitude of glycans that express (multi-)fucosylated terminal LDN and LeX motifs highly expressed by cercarial and egg-derived glycans. IgG response is highest when egg output starts to decline. We conclude that anti-glycan antibody dynamics are associated with the elimination of Schistosoma infection by rhesus macaques. Whether these anti-glycan responses are the cause or result of disease clearance will be further discussed.

Does the managemental conditions influence coccidiosis in commercial chicken? (P171)

Raman Muthusamy, Thenmozhi V, Raman M, Gomathinayagam S, Blake D and Tomley F
Department of Veterinary Parasitology, Madras Veterinary College, Chennai, 600007 Tamil Nadu, India

Coccidiosis, caused by species of the apicomplexan parasite *Eimeria*, is an economically important disease of chickens. *Eimeria* species are present world-wide, and are ubiquitous under intensive farming methods and the prevalence of *Eimeria* species varies across different production systems. As intensive farming system such as Deep Litter and Elevated cage found to influence the prevalence of coccidiosis, a study was carried out in six states of South India in Commercial broiler, Commercial Layer and Colour Broiler system of management. Faecal samples were collected across each poultry unit following a 'W' shaped sampling pathway, collecting one fresh dropping every two to five paces. The species prevalence of *Eimeria* were arrived based on ITS-1 nested PCR assay. Of the samples collected (N=254) in Southern India, 158 (62.02%) were positive in ITS 1 nested PCR and the species such as *E.acervulina, E.maxima, E.necatrix, E.mitis* and *E.tenella* were identified. *E. tenella* was the most predominant species (61.95%), followed by *E. mitis* (36.4%) and *E. acervulina* (24.71%), *E. necatrix* (15.57%) and *E. maxima* (13.39%). The study indicated the precominance of *Eimeria tenella* in all the managemental practices. Surprisingly, less pathogenic and highly neglected *E. mitis* was the second most prevalent species in all the management system of practice.

Impact of Sex and Strains of Mice on Susceptibility to *Eimeria papillata* Infection (P172)

Mohamed A. Dkhil1,2, Saleh Al-Quraishy1, Mahmoud S. Metwaly1, Denis Delic3
1 Department of Zoology, College of Science, King Saud University, Riyadh, Saudi Arabia, 2 Department of Zoology and Entomology, Faculty of Science, Helwan University, Cairo, Egypt, 3 Institute of Molecular Parasitology, Heinrich-Heine University, Duesseldorf, Germany

This study aimed to investigate the impact of sex and strains of mice on susceptibility to *Eimeria papillata* infection. C57BL/6 (B6) and Swiss Albino (SW) mice were infected with *Eimeria papillata*. B6 mice appeared to harbor higher parasites than did SW mice. Infection with 1000 sporulated oocysts results in a high number of oocysts output in the faeces of mice of both strains. The course of infection is characterized by a maximum increase of the released oocysts at day 5 p.i. Male B6 mice were more susceptible than female ones to *E. papillata* infection. Also, male SW mice are more susceptible than female ones. Moreover, Alcian blue staining of the mice jejunum showed that, in both sexes and strains of mice, the difference in the number of goblet cells in the jejunal villi was significant ($p \leq 0.01$). Male B6 mice appeared with more goblet cells than females. Also, male B6 mice have more goblet cells than male SW mice. Generally, the jejunal levels of hydrogen peroxide, nitric oxide and malondialdehyde were higher in males than females and these levels were much expressed in SW mice than B6 mice. In addition, TNF-α mRNA and iNOs mRNA were more upregulated in B6 mice than SW mice and in males than females. Essential for the outcome of coccidial infection is the sex and the host strain, with males in general being more susceptible than females towards infection caused by *E. papillata.*

The potential impact of salmon migration patterns on *Gyrodactylus salaris* (P173)

Nicola McPherson1,2, Rachel Norman2, Chris Williams3, Nick Taylor1
1 Cefas laboratory, Barrack Road, Weymouth DT4 8UB; 2 University of Stirling, Stirling, FK9 4LA; 3 Environment Agency, Brampton, Huntingdon PE28 4NE

The monogenean ectoparasite *Gyrodactylus salaris* has devastated Atlantic salmon (*Salmo salar*) stocks in Norway since the 1970s. While the UK remains free of this parasite, it has been the subject of much research due to the scale of its potential impact. Differences in environmental conditions in UK rivers compared to Norway are likely to affect both the

parasite and its host. Research has shown that strains of salmon found in the UK are susceptible to the parasite, and that the higher temperatures in UK waters may increase the time to first birth in the parasite, but that this may be counteracted by an overall reduction in the parasites fecundity. The differences in temperature also lead to substantial differences in the salmon life-cycle timings between the two countries. For example salmon in the UK typically remain in freshwater for around two years, compared to four years in Norwegian rivers. As the parasite is only able to survive in freshwater, and consequently those attached to salmon smolts will die when their hosts migrate to sea, it is possible that this characteristic may alter the outcome of an outbreak in the UK, should an introduction occur. In order to determine the impact that salmon migration patterns may have on an outbreak of *G. salaris*, an existing macroparasite model was further developed to incorporate seasonality and the anadromous life cycle of salmon. Preliminary results are presented along with plans for future work.

Prevalence of *T. gondii* in pigs from Yucatan (P174)*

Ana Isabel Cubas Atienzar1, Matilde Jiménez-Coello2, Antonio Ortega-Pacheco2 and Judith E. Smith1.
1 *University of Salford, 2 Universidad Autónoma de Yucatán*

Toxoplasmosis is a worldwide distributed zoonotic parasitic disease caused by *Toxoplasma gondii*. Humans can become infected mainly by ingestion of infected meat and by contaminated water and vegetables. As pork is one of the most highly consumed meats, pigs are an important source of human infection. The prevalence of *T. gondii* was assessed on two intensive farms located in Yucatan, in Mexico. Serum IgG antibodies levels were measured in 53 fattening pigs with an indirect ELISA kit (Human-GmbH, Wiesbaden, Germany) and the target B1 gene was amplified by PCR in tissues samples for 11 individuals. Isolation of *T. gondii* also was attempted from tongues and/or blood samples of 13 animals. Questionnaire information was collected of the farm and animals characteristics and they were studied as a possible risk factors of *T. gondii* infection. Results of this preliminary survey showed a high prevalence (85%) of *T. gondii* antibodies and some evidence of parasites in tissue. Both farms were positive for *T. gondii* infection and prevalence increased with the age. In terms of possible transmission networks the number of cats was high on both farms and both also had bird and rodents access, and the feeders were open to the environment. Later sampling of cats from one farm led to isolate *T. gondii* by mice bioassay.

Mother's secret recipe: variation in *trans*-generational immune priming investment in *Tenebrio molitor* (P175)*

Charlotte Miller
School of Biological Sciences, Queen's University, Belfast

Trans-generational immune priming (such as the transfer of antibodies from mother to offspring in vertebrates) allows offspring to benefit from the immune experience and resources of their parents, giving them a competitive advantage in an infected population. In insects, the process incurs costs associated with immune activation for both the parents and the offspring, making its adaptive value highly variable, and clarifying why only infected mothers prime their offspring. In this study, female *Tenebrio molitor* were injected with varying concentrations of an immune elicitor and allowed to reproduce with naïve males. Offspring levels of the pro-phenoloxidase peptide, which is activated to make the immune enzyme phenoloxidase, were positively correlated with the concentration of elicitor the mother was injected with. This relationship was shown in offspring which received a standard challenge across maternal treatment groups and also unchallenged offspring. Phenoloxidase levels were unaffected by the maternal treatment. The proportional response in the pro-phenoloxidase peptide allows an increase in the constitutive defence of offspring which is fast-acting and broadly effective but less costly than other mechanisms. Pro-phenoloxidase level variation as a priming mechanism has not been previously documented, and is not commonly tested for when investigating the priming of insect offspring by mothers. This study demonstrates the necessity of cataloguing the effects of trans-generational immune priming across the immune system.

The perfect burrow - predicting biological conditions required for plague (*Yersinia pestis*) in the Pre-Balkash desert, Kazakhstan (P176)*

Bethany Levick, Anne Laudisoit, Mike Begon
Institute of Integrative Biology University of Liverpool Biosciences Building Crown Street, Liverpool, L69 7ZB

Now confined to distinct wildlife foci across the world, the historical impact of plague is demonstrative of its potential for devastation. As the risk of zoonotic diseases such as plague crossing over into human populations increases, it is vital

that we are able to minimise the opportunity for this to occur and the risk to human populations if this does happen. The plague focus in Kazakhstan has been studied for around 15 years, and a mathematical model for predicting plague in this area currently exists. This predicts plague will occur above a minimum curved threshold generated as a function of gerbil density (*Rhombomys opimus*) and flea burden(typially *Xeopsylla* spp.), arising through plague spreading through the gerbil population as a percolation phenomenon. Comparison of the predictions generated by this model with field data finds that plague is never found where biological conditions fall below the threshold, but that in some instances conditions that are above the threshold are observed where plague is not present. To investigate further environmental factors controlling the plague system data collected across the Pre-Balkash area between 2010 and 2013 was analysed to find environmental factors predictive of burrow occupancy and flea burden. This initial analysis has identified both gerbils and the flea burden can be predicted by a number of landscape and environmental properties of the surrounding area, with some shared predictors. These relationships should allow us to use data collected in the field to make predictions about where plague is then likely to occur.

Leucine aminopeptidases in *Trypanosoma brucei* (P177)

Priscila Pena-Diaz, Chris Resl, Pavel Flegontov, Olga Flegontova, Julius Lukes
Institute of Parasitology, Biology Centre, Branišovska 31, 370 05 České Budějovice, Czech Republic

LAP3 are a group of leucine aminopeptidases of the M17 family, ubiquitous in cellular systems. These proteins have been found to be involved in a variety of moonlighting functions, such as transcription regulators, recycling glutathione in the gamma-glutamyl cycle, regulating meiosis, interacting with Fe-S clusters proteins and acting as chaperones in stress conditions. In *Trypanosoma brucei*, there are three LAP3 homologues –described as a, b and c, and their transcripts are amongst the most abundantly expressed in the cell. Depletion by RNAi in the procyclic form has a moderate effect on cell viability, which may imply redundancy. The effect of the depletion by RNAi in the cell is increased in the presence of H_2O_2. The proteins were tagged, both endogenously and for overexpression, and their localisation was assessed by immunofluorescence. They were found to be expressed in different compartments of the cell; with LAP3a being associated to mitochondrial DNA, LAP3b locating to the nucleus, and LAP3c as a cytosolic protein. Over-expression of both LAP3a and LAP3b seem to have an adverse effect on cell viability, possible implying a possible regulatory effect. The implications of the differential compartmentalisation and redundancy are discussed.

Ultra-high throughput screening for anti-Wolbachia drugs to treat Onchocerciasis and Lymphatic Filariasis (P178)*

Rachel Clare, Roger Clark, Kelly L. Johnston, Darren A. Cook, Louise Ford, David C Murray, Eileen McCall, Kirsty Rich, Stephen A. Ward, Mark J. Taylor
Department of Parasitology, Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA, UK; AstraZeneca, Discovery Sciences, Alderley Park, Cheshire, SK10 4TG, UK

The Anti-Wolbachia consortium (A•WOL) at Liverpool School of Tropical Medicine (LSTM) has partnered with the Global High Throughput Screening (HTS) Centre at AstraZeneca(AZ) in the first open access HTS project for the World Intellectual Property Organization's (WIPO) Re:Search program against Neglected Tropical Diseases. The A•WOL consortium aims to identify novel macrofilaricidal drugs targeting the essential bacterial symbiont (Wolbachia) of the filarial nematodes causing onchocerciasis and lymphatic filariasis. The project will scale-up the throughput of the A•WOL cell-based screening assay using AZ's leading automation, screening technologies and expertise. A•WOL currently use a Wolbachia infected C6/36 *Aedes albopictus* cell line imaging assay (running on the Perkin Elmer Operetta® platform) to screen up to 10x 384 well plates per day. This assay uses texture analysis of cells stained with SYTO®11 (fluorescent DNA stain) as a direct measure of bacterial load. The first step in the assay scale up process is to validate the use of assay ready cryopreserved cells for direct transfer into compound plates. The use of a single cell batch, should simplify the screening process at scale, whilst reducing variation over the screen. The assay is also being modified to optimise antibody based techniques compatible with the TTP Acumen® (Laser scanning cytometer) or Perkin Elmer EnVision® (whole well fluorescence) plate readers. Subject to further optimisation and validation, this assay will enable screening against AZ's full chemical library of compounds in a single screening activity. Hits identified will then be progressed through the A•WOL drug discovery and development programme for new macrofilaricides.

***Trypanosoma brucei* Nfu proteins (P179)**

Corinna Benz, Corinna Benz, Julie Kovářová, Julius Lukeš
Institute of Parasitology, Biology Center and Faculty of Sciences, University of South Bohemia, České Budějovice, Czech

Republic

Iron-sulphur clusters (ISCs) are important co-factors for numerous proteins with important cellular functions including metabolic catalysis, DNA replication and repair, iron regulation and protein translation. The core ISC assembly machinery is found in the mitochondrion, where several target proteins also reside. For the generation of cytosolic and nuclear iron-sulphur (Fe/S) proteins a still unknown compound generated within the mitochondrion is exported to the cytosol, where it is utilised by the cytosolic iron-sulfur protein assembly machinery (CIA) to generate ISCs. All ISCs are eventually transferred to specific apoproteins by ISC targeting factors. Nfu1, which is present in all eukaryotes, is one of these targeting factors generally found within the mitochondrion. In humans, Nfu1 is required for proper assembly of a small subset of [4Fe-4S] proteins, including subunits of respiratory complexes I and II and lipoic acid synthase. Whereas most eukaryotes harbour a single Nfu gene, plants and trypanosomatids have five and three respectively. The function and localisation of Nfu1, Nfu2 and Nfu3 was analysed in procyclic form *Trypanosoma brucei* and results will be presented.

Drug Development for Chagas Disease (P180)

Lorna MacLean, John Thomas, Manu De Rycker, David Gray
Drug Discovery Unit, Division of Biological Chemistry and Drug Discovery, College of Life Sciences, University of Dundee, DD1 5EH, UK

Chagas disease is caused by the protozoan parasite *Trypanosoma cruzi* which is mainly transmitted to humans by the faeces of triatomine bugs. An estimated 7-8 million people are currently infected mostly in Latin America, however, increasingly cases are being diagnosed worldwide. *T. cruzi* infection can lead to cardiac, digestive and neurological disorders in the chronic phase resulting in 12,000 deaths annually. The current treatments (Benznidazole & Nifurtimox) are inadequate providing limited rate of cure in chronic cases and drug resistance has been reported, therefore, there is an urgent need to develop new drugs. As part of our neglected diseases drug discovery programme at the DDU we have developed a suite of assays for *T. cruzi* hit discovery. Our screening cascade is designed to combine high-throughput and robustness with *in vivo* predictivity. Our main screening platform is a high-content image-based intracellular assay. Hit-rates against *T. cruzi* tend to be high so we perform tri-point and potency follow-up assays to select the most promising compounds which are then tested for cidality in a rate of kill assay. This cascade has successfully identified 3 series with *in vivo* activity in an acute model of Chagas disease. We are currently developing a panel of *T. cruzi* isolated from patients encompassing discrete typing unit (DTU) I, III, IV, V and VI to verify cross-strain activity for lead-op candidates.

Characterization of the cell surface proteome of the plant parasite *Phytomonas serpens* (P181)*

Eleanor Ceindeg Jaskowska, Steve Kelly
Department of Plant Science, University of Oxford South Parks Road Oxford OX1 3RB

Phytomonas, a sub-group of trypanosomatid parasites, are spread between plant hosts by biting insects. *Phytomonas* species are globally distributed and are found routinely in in some of the world's most important crops including coffee, cassava and oil palm. While some species have been reported to cause disease symptoms, the majority of species cause no detectable effect on their plant hosts, therefore *Phytomonas* have attracted little attention from the scientific community. While the mechanism(s) by which *Phytomonas* parasites evades plant immune responses is unknown, they share the same origin of parasitism with mammalian pathogens *Leishmania* and *Trypanosoma*. Thus adaptation of an ancestral protein cohort, likely involving cell-surface proteins, probably facilitated parasitism of plants. Here, we develop an optimised cell surface protein isolation protocol and use it to characterize the cell surface proteome of the non-pathogenic tomato parasite *Phytomonas serpens*. Proteins identified by this technique include the homolog of KHARON1 which is localised to the flagellar pocket in *Leishmania* and is involved in trafficking proteins to the flagellum. This novel proteome sheds light on the evolution of parasite cell-surfaces within this globally important group.

Pyrimidine nucleoside transporters in *L. major* and *L. mexicana* (P182)*

Khalid Jamaan H Alzahrani1,2, Juma A. M. Ali1,3, Harry P. de Koning1
1 Institute of Infection, Immunity and Inflammation, University of Glasgow, Glasgow, United Kingdom, 2 Faculty of Applied medical Sciences, Taif University, Taif, Saud Arabia, 3 Faculty of Science, Al Jabal Al Gharbi University, Gharyan, Libya,

Research has delineated four nucleobase and nucleoside transporters in *Leishmania* promastigotes, designated as NT1-4

with the vast majority of studies on NT1 and NT2 being carried out using *L. donovani* genes. However, it is not known whether the same genes mediate purine/pyrimidine transport in other *Leishmania* species. In this study, we evaluated the uptake of ^{3}H-uridine and ^{3}H-thymidine in *L. major* and *L. mexicana* promastigotes. The uptake of both nucleosides was mediated by LmexNT1, and LmajNT1, respectively. *L. mexicana* NT1 showed similarly efficient transporter of uridine (Km =7.2±0.9 µM and Vmax =0.33±0.11 pmol·107 cells-1·s-1) and thymidine (Km =4.2±0.4 µM and Vmax =0.85±0.12 pmol·107 cells-1·s-1). Thymidine (Ki = 14.2±3.1) and adenosine (Ki = 0.23±0.04) significantly inhibited uridine uptake. Transport of ^{3}H-thymidine was also inhibited by uridine and adenosine with Ki values of 6.0±0.6 µM and 0.25±0.04 µM, respectively. In *L. major*, LmajNT1 displayed high affinity for uridine, with a Km value of 7.3±1.6 µM and Vmax of 0.078±0.005 pmol·107 cells-1·s-1, which was inhibited by thymidine, 2'deoxyuridine and adenosine. ^{3}H-thymidine uptake showed a Km value of 30.7±2.1 and Vmax of 0.14±0.09, and was inhibited by uridine and adenosine. These observations are all consistent with the *L. donovani* model for nucleoside transport being conserved in *L. major* and *L. mexicana*. We have now expressed *L. mexicana* and *L. major* NT1.1, NT1.2 and NT2 in a *T. b. brucei* strain lacking the aminopurine transporter TbAT1, in order to confirm the genetic identities of the transport activities observed in promastigotes.

Immune Variation in a Wild Mammal Population (P183)

Rebecca Watson, Dan Nussey1, Rose Zamoyska1, Tom McNeilly2
1 University of Edinburgh, 2 Moredun Research Institute

The evolutionary and ecological context of age-related immune decline has been little studied in free-living animals, and yet could offer great insight into the interactions between immune phenotype and environment. Wild populations, in which individuals experience a harsh environment and face simultaneous natural infections, provide important insights into how natural selection has shaped variation in immune responses. In this study we measured a range of immune markers, including proportions of different leukocyte and T cell sub-populations, in a population of wild Soay sheep living on the St. Kilda archipelago. Our study population has been the subject of long term study and is an ideal opportunity for longitudinal data collection and analysis. We are able to measure individuals over consecutive years and make comparisons both within and between individuals, age classes and the sexes. Here we test for changes in proportions of T cell sub populations with age and relate these changes to trends in other life history components, including survival and reproduction. We also test for immune variation between the sexes. The relationships demonstrate the evolutionary impact of the natural environment and life history trade-offs on immune function variation between individuals, as well as within overall population dynamics.

A novel method to regulate expression of organellar proteins in *Leishmania major* using the destabilisation domain (P184)

David Wildridge, Brian Panicucci, Michaela Veselíková, Alena Zíková
Laboratory of Functional Genomics of Protists Department of Molecular Parasitology Institute of Parasitology Biology Centre, ASCR, v.v.i. Branišovská 31 370 05 České Budějovice, Czech Republic

The investigation of essential genes in *Leishmania major* has proved challenging due to the lack of RNAi machinery and a regulatable gene expression system. Alternatively, protein expression can be controlled through the regulated degradation of a target protein fused to the FK506/rapamycin-binding protein destabilisation domain (DD). In the presence of the stabilising ligand FK506, the DD is properly folded and confers stability of the target protein that would otherwise be degraded. This has successfully been used to investigate several cytosolic and nuclear proteins in *T. gondii, P. falciparum, L. major,* and *L. braziliensis*, although regulation is reported to be protein dependent. Our attempts to regulate proteins targeted to the mitochondrion were unsuccessful. Whilst stabilisation of the target protein was possible through the addition of FK506, subcellular fractionation experiments revealed that the protein was only localised to the cytosol. Consequently, we developed a method that utilises the bacteriophage T7 RNA polymerase fused to the DD to regulate expression of a gene of interest under the control of a T7 promoter at a transcriptionally silent locus. This would allow us to knockout the endogenous alleles of essential genes. The major advantage of this approach is that stabilisation of the T7 RNA polymerase is independent of the protein of interest, thus allowing the investigation of organellar proteins. Our preliminary data indicates that levels of T7 RNA polymerase can be regulated in an acceptable time-dependent manner and we are assessing the feasibility of this cell line to generate regulatable knockout cell lines.

Metabolomics & Modelling: deciphering the interplay between energy metabolism and oxidative stress response in bloodstream form *Trypanosoma brucei* (P185)

Fiona Achcar, Dong-Hyun Kim, Eduard J. Kerkhoven, Darren J. Creek, Barbara M. Baker, Rainer Breitling, Michael P. Barrett
University of Glasgow

Human African Trypanosomiasis is a potentially lethal disease caused by the protozoan parasite *Trypanosoma brucei*. The metabolism of the bloodstream form of the parasite has several unique features that have been investigated, in the search for potential drug targets. Mathematical modelling has been used as a valuable tool to decipher glycolysis, the parasite's only energy source, since 1997. Recently, we extended this model of glycolysis to include the pentose phosphate pathway, another essential pathway that generates the NADPH used in the cells' protection against oxidative stress, and thus provides a metabolic link to another important drug target in trypanosomes. Mass spectrometry based metabolomics is another valuable tool that enables us to gain a deeper understanding of the parasite's metabolism, either by comparing the metabolic state of cell grown in two conditions or by using labelled precursors to follow metabolic pathways. Here, we combine modelling and metabolomics to investigate further the link between metabolism and oxidative stress response in trypanosomes.

Praziquantel Reverses Schistosomiasis-induced Pulmonary Arterial Hypertension and Vascular Remodeling (P186)

Alexi Crosby1, Frances M. Jones2, Ewa Kolosionek3, Mark Southwood1, Ian Purvis1, Elaine Soon1, Ghazwan Butrous3, David W. Dunne2, Nicholas W. Morrell1
1 Department of Medicine, Cambridge University, UK, 2 Department of Pathology, Cambridge University, UK, 3 University of Kent, UK

Schistosomiasis is the most common world-wide cause of pulmonary arterial hypertension (PH). We sought to determine whether praziquantel could reverse established pulmonary vascular remodelling and PH in a mouse model of *Schistosoma mansoni*. Mice were infected percutaneously with *Schistosoma mansoni*. 17-weeks post-infection mice were sacrificed or received two doses of praziquantel by oral gavage or vehicle. At 17 or 25 weeks post-infection right ventricular systolic pressure (RVSP) and the degree of right ventricular hypertrophy (RVH) (as markers of PH) were measured. The liver and lungs were removed for egg counts and histology. Serum and lung cytokine expression were determined. Infected vehicle treated mice demonstrated significant increases in RVSP and RVH at 25 weeks, which was highly dependent on the presence of eggs in the lung. The increase in pressure was accompanied by dramatic pulmonary vascular remodelling of vessels in close proximity to eggs and an increase in serum and lung cytokines. Praziquantel led to clearance of eggs from the lung and the liver. Praziquantel prevented the rise in RVSP, RVH, pulmonary vascular remodelling and lung cytokine expression. This is the first study to show an increase in RVSP and RVH following chronic infection with *S. mansoni*, validating this as a model of PH. The increase in RVSP was dependent on the presence of eggs in the lung and is likely to be due to the increase in local lung cytokine levels see in these animals. The progression of disease can be reversed by praziquantel.

A toolkit enabling efficient, scalable and reproducible gene tagging in trypanosomatids (P187)

J Sunter, S Dean, R Wheeler, I Hodkinson, Eva Gluenz, K Gull
Sir William Dunn School of Pathology, University of Oxford Department of Computing, Imperial College London

The development of high throughput methods to analyse the DNA, RNA and protein content of cells has created large datasets that contain numerous proteins of interest. A common first step in analysing protein function is to determine protein localisation using a fluorescent tag; however, the methods for tagging proteins have not kept pace with the developments in genomics, transcriptomics and proteomics, creating a bottleneck in the analysis of these large datasets. We have developed rapid, high throughput, PCR based tools for endogenous tagging of proteins in trypanosomatids. We generated a single plasmid that when coupled with long primer PCR can be used to create constructs for both N and C-terminal endogenous eYFP tagging of proteins in procyclic and bloodstream form *Trypanosoma brucei*. Importantly, we optimised the workflow to make it efficient, scalable and reproducible. This long primer PCR approach is not effective in *Leishmania mexicana*. Our studies show that a likely explanation is that transfection efficiency was proportional to the length of homology between the tagging construct and the target gene; efficient transfection could only be achieved with longer homologous regions than can be generated using long primer PCR. Hence, we developed a fusion PCR based approach to generate sufficiently long homologous regions to create endogenous tagging constructs, suitable for use in trypanosomatid species with less efficient homologous recombination. This again has great benefits over existing procedures. Our set of tools for the endogenous tagging of proteins in trypanosomatids is efficient and will accelerate the analysis of protein localisation and function.

The role of dendritic cells in inducing Th2 responses to *Schistosoma mansoni* eggs (P188)*

Johannes U. Mayer, Lauren Webb, Stephanie Houston, Vuk Cerovic, Andrew MacDonald, Simon Milling
Institute of Infection, Immunity and Inflammation College of Medical, Veterinary and Life Sciences University of Glasgow Sir Graeme Davies Building 120 University Place Glasgow G12 8TA United Kingdom and Faculty of Life Sciences Core Technology Facility 46 Grafton Street Manchester M13 9NT United Kingdom

Schistosoma mansoni eggs (SME), which are the main cause of chronic Schistosomiasis, provoke a strong Th2 immune response that can lead to granulomatous reactions and fibrosis in affected organs. Recently, CD11c depletion has been shown to severely disrupt Th2 immune responses against *Schistosoma mansoni*, suggesting that antigen presenting cells, which express CD11c, mediate this immune response. Dendritic cells (DCs) are the most likely CD11c-positive candidates, as they continuously migrate from epithelia, carrying antigens, to the draining lymph nodes where they activate naive T-cells. However, the precise role of DCs in inducing immune responses against SME have not been studied. We have developed an *in vivo* mouse model where we inject SME in the subserosal layer of the small intestine of C57BL/6 mice, the anatomical location where SME naturally become lodged. This provokes a Th2 response in the draining mesenteric lymph nodes (MLNs). To test the role of DCs in this response, we will purify migrating DCs from egg challenged mice and control animals and then inject these cells into the subcapsular region of the MLN of naive mice. Detection of an antigen-specific Th2 response in those mice will indicate that migratory DCs are sufficient to carry and present egg antigens and drive Th2 responses in our model. This knowledge will contribute to a better understanding of how the early immune response against SME is mediated. This information can be applied to battle chronic Schistosomiasis and may help understand Th2 driven responses against other parasites or Th2 allergic responses.

A Novel Cell-Based Screen for Discovery of a Macrofilaricide (P189)

Vera Unwin, Vera Unwin, Kelly L. Johnston, Mark J. Taylor
Department of Parasitology, Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA, UK

The drug discovery pipeline for a macrofilaricide has been hampered by the complexities of the filarial nematode life-cycle. Current *in vitro* screening relies on cumbersome, low-throughput microscopy-based assays, which are ultimately dependent upon the provision of adult worms from animal reservoirs. With a Bill & Melinda Gates Foundation Grand Challenge Exploration award, we aim to circumvent this barrier by developing a robust filarial nematode cell line that can be used in a high-throughput screening (HTS) assay. Our novel approach is to develop “micro-cultures” of cells seeded on microtitre plates and use the automated Operetta High Content Imaging system (Perkin Elmer) to monitor their growth dynamics. Using this platform for the development of the cell line will help to maximise throughput of resulting drug screening assays and also identify the optimum culture conditions. These conditions, including culture medium, supplementation, temperature and the techniques/ timeframes for passage are being assessed by parameters such as cell viability, morphology, proliferation and differentiation of each culture using the Operetta High Content Imaging system. This will allow us to select the most promising cultures, maximising the chance of establishing a robust filarial cell line. Sourcing cells from *Brugia malayi* due to its more extensive genetic characterisation, we aim to characterise the resulting cell line using transcriptomic and proteomic analysis, as well as phenotypic data acquired using the Operetta. Such a cell line would become a potentially valuable tool for drug discovery, as well as for broader studies on the cell biology of these parasites.

A Genome on the Verge of Extinction (P190)

Caroline Durrant1, Nancy Holroyd1, Matthew Berriman1, Mark L. Eberhard2, Ernesto Ruiz-Tiben3, James A. Cotton1
1 The Wellcome Trust Sanger Institute, Hinxton, UK, 2 Division of Parasitic Diseases and Malaria, Centers for Disease Control and Prevention, Atlanta, Georgia, 3 The Carter Center, Atlanta, Georgia

Dracunculus medinensis (Guinea worm) is a nematode that causes the disease dracunculiasis. This was once a major parasitic infection, and widespread across tropical Africa and Asia. However, infection is entirely preventable, through provision of clean water and behaviour change, which has led to a drop in cases from around 3.5 million cases per annum in the 1980s to fewer than 150 cases in 2013. The disease is now eradicated in all but four countries. The aim is that dracunculiasis will be the second human disease to be eradicated, the first parasitic disease and the first disease to be eradicated without the use of a vaccine or drug. However, dracunculiasis was believed to be extinct for a decade in Chad until new cases emerged in 2010. The pattern of the current outbreak in Chad does not cluster by village or water

source as expected and a large number of dogs were also infected. It appears that dogs are now the main host in Chad, with human cases being sporadic and incidental, transmitted by a common paratenic host. Building on de novo genome assemblies for *D. medinensis* and the related *D. insignis*, we describe genome-wide patterns of diversity in *D. medinensis* samples from Chad and from other parts of the range. We are aiming to understand both the current diversity of the population in endemic countries and the diversity of populations before the eradication campaign, and to more formally understand the epidemiology of canine and human cases in Chad.

Investigating dendritic cell subsets during Th2 induction against *Schistosoma mansoni* infection (P191)*

Angela Marley1, Alex Phythian-Adams1, Peter Cook1, Gareth-Rhys Jones1, Lucy Jones2, Lauren Webb1, Rick Maizels2, Andrew MacDonald1

1 Manchester Collaborative Centre for Inflammation Research, University of Manchester, M13 9NT, UK, 2 Institute of Immunology and Infection Research, School of Biological Sciences, Ashworth Laboratories, University of Edinburgh, EH9 3JT, UK

In *Schistosoma mansoni* infection T cells are directed towards strong T helper type 2 (Th2) responses that are crucial for the survival of the host. Dendritic cells, a heterogeneous population of innate antigen presenting cells, provide an essential link between innate and adaptive immunity and are important for the induction and direction of T cell responses. Using a mouse model for the human disease we have previously shown that the induction of Th2 responses in *S. mansoni* infection is dependent on the presence of CD11c+ Dendritic cells (DC). However, the DC subsets involved and the precise mechanisms they employ to provoke Th2 responses remain poorly defined. To progress our understanding of which DC subsets are important for this process we have used Batf3KO mice, which are deficient in CD8α+ DCs and, in some tissues, migratory CD103+ DCs. CD8α+ DCs are most commonly associated with generation of Th1 immunity against bacterial and viral infections, and Ag cross-presentation, but their role in helminth infection and Th2 settings is currently poorly understood. We have shown for the first time that CD8α+ DCs are not essential for Th2 induction during helminth infection. However, Batf3KO mice infected with *S. mansoni* displayed dysregulated Th2 responses in the absence of CD8α+ DCs. Ongoing work aims to further understand this important and previously unappreciated role for CD8α+ DCs in orchestration of the immune response during helminth infection.

Cold Blooded Parasitism! Molecular identification of *Hepatozoon pettiti* (Hoare, 1932) and its evolutionary interactions with the Nile crocodiles (*Crocodylus niloticus*) of the Okavango Delta (P192)

Scott P. Lawton1, Polly M. Hayes1,2, Amy Stupartis1, Narayan S. Suman1, Alison J. Leslie3, Nico J. Smit4, Angela J. Davies1

1 Kingston University London, Kingston Upon Thames, UK, 2 Natural History Museum, London, UK, 3 Stellenbosch University, South Africa, 4 North West University, Potchefstroom, South Africa

Hepatozoon pettiti is an Apicomplexan intraerythrocytic parasite of the Nile crocodile (*Crocodylus niloticus*) with high prevalence throughout Africa and is transmitted by tsetse flies, although leeches are suspected to also play a role in transmission. The aims of this study were to firstly identify *H. pettiti* in blood fed leeches using 18s rDNA sequences and match them with parasites in blood films from the crocodiles, and secondly to understand the evolutionary interactions between *H. pettiti* and the crocodiles of the Okavango by measuring variation in the major histocompatibility complex (MHC) II b gene of both infected and uninfected individuals. DNA was extracted from archived material from a previous study in 2006 which included leeches preserved in ethanol and blood films from the crocodile hosts. Hepatozoon 18s rDNA sequences from leech blood meals were obtained and were successfully matched with corresponding blood films from infected crocodiles. Also a small fragment of MHC was sequenced from the blood films and specific genotypes that appeared to be more susceptible to the parasite than others were observed. There was reduced variation in infected MHC genotypes potentially indicating that parasites are adapting to the most common genotypes within a genetically diverse crocodile population. This is of considerable concern as Nile crocodiles that are being commercially bred are shown to have a substantially genetic variation which could lead to parasites such as *H. pettiti* becoming highly pathogenic and causing considerable economic losses as an emerging disease.

Revealing the mechanisms of benznidazole-resistance in *Trypanosoma cruzi* (P193)

Ana M. Mejía-Jaramillo1, Laura González1, Paola García-Huertas1, Andres Felipe Diez1, Maria C. Echeverry2, John Kelly3, Omar Triana-Chávez1,

1 Biología y Control de enfermedades Infecciosas-BCEI, Universidad de Antioquia, 2 Departamento de Salud Pública,

Facultad de Medicina, Universidad Nacional-Sede Bogotá, 3 London School of Hygiene and Tropical Medicine

Benznidazole (Bz) and Nifurtimox (Nfx) are the first choice treatment for clinical management of acute stage Chagas disease. These are pro-drugs that need to be activated within the parasite by an NADH-dependent type-I nitroreductase (TcNTR). Resistance of *Trypanosoma cruzi* to these compounds can be mediated by alterations in the TcNTR gene copy number and/or by mutations that disrupt enzymatic function. However, evidence shows that the wide diversity of Bz-susceptibility *in T. cruzi* isolates from the field need not be associated with alterations in the TcNTR sequence and that laboratory-generated resistant clones can display different levels of cross-resistance to Bz and Nfx. It is implicit therefore that mechanisms linked to TcNTR activity alone are insufficient to account for the wide range of Bz-resistance phenotypes observed in both the laboratory and the field. The present study aimed to identify new routes to Bz-resistance in *T. cruzi*. Using high-throughput approaches, we compared the transcriptome and the proteome of Bz-sensitive and resistant *T. cruzi* clones from a paediatric Colombian patient. We found 22 up-regulated and 29 down-regulated transcripts associated with resistance, and 19 proteins that were differentially expressed. Six of these gene products were identified by both approaches. Eight had previously been reported as resistance-associated proteins. Further studies to evaluate the possible role of these genes in conferring Bz-resistance have been performed by over-expression of some of the proteins in *T. cruzi* and knockdown of the orthologous genes in *Trypanosoma brucei*.

Akt signalling in the human parasite *Schistosoma mansoni*. (P194)*

Maxine Mckenzie, Maxine Mckenzie, Ruth S Kirk, Anthony J Walker
School of Life Sciences, Kingston University, KT1 2EE, UK

Protein kinases are intracellular cell signalling enzymes that co-ordinate cellular function. Although the human parasite *Schistosoma mansoni* is known to contain at least 252 protein kinases, we know very little about their mechanisms of activation and downstream functional responses. Our research on *S. mansoni* has therefore begun to focus on the Akt signalling pathway, which in humans is regulated by phosphoinositide 3-kinase (PI3K) and is implicated in processes such as cancer progression, insulin signalling, glucose metabolism and transcriptional regulation. Western blotting of serum treated 24 hour *S. mansoni* schistosomules with anti-phospho Akt (Thr 308 or Tyr 315) antibodies that recognize the phosphorylated (activated) form of the enzyme revealed that activated Akt is present and has an approximate apparent molecular mass of 55 kDa. Moreover, the phosphorylation sites Thr 308 and Tyr 315 of human Akt were well conserved in *S. mansoni* Akt. Immunoprecipitation of phosphorylated Akt from protein extracts of serum treated adult worms and schistosomules with anti-phospho Akt (Thr 308) antibodies demonstrated that the immunoreactive Akt possessed kinase activity towards the downstream substrate glycogen synthase kinase 3 (GSK-3) in both life stages with the schistosomules displaying considerably more active Akt than adult worms. Akt might therefore play a more prominent functional role in developing schistosomules than in mature adult worms.

Evaluating the trypanocidal activity of quinone-based compounds (P195)*

Emma Louise Meredith1, Sam Alsford2, Shane R Wilkinson1
1 School of Biological and Chemical Sciences, Queen Mary University of London, Mile End Road, London E1 4NS, 2 London School of Hygiene and Tropical Medicine, Kepper St, London, WC1E 7HT

Quinones are a group of molecules characterised by the presence of two carbonyl groups linked to a carbocylic backbone. Some of these compounds are of medicinal value functioning as inhibitors of oxidoreductase pathways or as prodrugs. Here, we show that a series of 1,4-benzoquinones, particularly aziridinyl-1,4-benzoquinones (ABQs), display potent anti-parasitic activity against bloodstream form *Trypanosoma brucei*. To understand their mechanism of action, RH1 the archetypal ABQ, was used to select for resistant *T. brucei* populations in a loss of function RNAi library screen. A low throughput, PCR-based approach demonstrated that the major transcript down-regulated in these cultures coded for a type I nitroreductase (TbNTR), confirming that this ABQ is a pro-drug. Functional studies using parasite lines expressing elevated levels of TbNTR confirmed the importance of NTR activity to not only RH1 but to most other ABQs tested. In a parallel, RH1 resistant *T. brucei* populations were selected in increasing concentrations of this ABQ and clonal lines (RH1R) generated. The resultant parasites exhibited a 3-fold increase in their IC50 to RH1 and displayed cross-resistance to other aziridinyl quinone/nitrobenzyl-based compounds and to nifurtimox. We postulate that resistance to RH1 in the selected line involves a common mechanism shared between all of these compounds, with this most likely to occur through reduction in the level of TbNTR-mediated pro-drug activation.

Modulation of chemokine production by African trypanosomes (P196)*

Edina Szabo, Jeremy M Sternberg, Donna M MacCallum
University of Aberdeen, School of Medical Sciences, Foresterhill, Aberdeen AB25 2ZD UK

African trypanosomes invade the blood stream and several internal organs in the early stage of infection and cross the blood-brain-barrier in late stage infections. Infection is accompanied by dysregulation of host immune responses. Host epithelial and endothelial cells, as well as immune cells, are able to respond to pathogens through cytokine and chemokine production, which suggest that non-immune cells may be involved in the initiation of immune responses during infections. As an example, interactions between parasites and thymic epithelial cells are important for effective host immune responses. Previous studies have demonstrated that *T. musculi* is able to stay cryptic in the kidneys with no induction of immune recognition. Therefore we have investigated renal epithelial cell responses to *T. brucei*, using a new *in vitro* model developed for the study of host-pathogen interactions. Our preliminary experiments showed that production of KC and MIP-2 were reduced in the presence of *T. brucei*, correlating with low levels of epithelial cell damage. This suggests that *T. brucei* able to suppress immune responses during infection, and therefore host epithelial/endothelial cells may play a role during dysregulation of host immune responses.

The impact of essential oils on *Culex pipiens* Larvae (Diptera, Culicidae)- *In vitro* assessment (P197)

Ahmed Abdelnabi, Rashed, A.A1&4. Hamada, M.S.2, Ostroff G.3, George D.1, Sparagano O.A.E.1
1 Faculty of Health and Life Sciences, Department of Applied Sciences, Northumbria University, Newcastle upon Tyne, UK, 2 Department of Pesticides, Faculty of Agriculture, Mansoura University, Mansoura, 35516, Egypt, 3 University of Massachusetts Medical School. Program in Molecular Medicine, Worcester, MA 01605, USA, 4 Economic Entomology Department, Faculty of Agriculture, Mansoura University, Mansoura, 35516, Egypt.
Egypt.

Mosquitoes are considered one of the most serious arthropod pests for humans due to the number of diseases they can transmit. The control of mosquitoes is an important public health concern around the world. *Culex pipiens* is considered the most common and widely distributed mosquito species across Egypt. In the current study *in vitro* mosquito larvicidal assays were conducted to determine the LC50 and LC95 values for a group of yeast particle encapsulated monoterpenoids (Citral, Geraniol and Thymol) against second, third and fourth instar larvae of *Culex pipiens*. LC values were determined after 24 and 48 hours. After 24 hours Thymol was most potent against the second, third and fourth instar larvae with an LC50 of 1.03 (LC95: 5.40), 1.74 (LC95: 3.45), 2.15 (LC95: 4.96) ppm, respectively. After 48 hours all of the tested encapsulated monoterpenoids exhibited strong efficiency against all larvae instars. The best essential compound against the second and the fourth instars was Citral with an LC50 value of 0.49 (LC95: 12.65) and 0.83 (LC95: 2.60) ppm, after 24 and 48 hours, respectively. Against third instar larvae Geraniol was the most potent terpene with an LC50 value of 0.83 ppm (LC95 = 3.19 ppm).

Transketolase in *Leishmania mexicana*: regulation of subcellular localisation and metabolic roles (P198)*

Julie Kovářová1, David Wildridge1, Fiona Achcar1, Frédéric Bringaud2, Michael Barrett1
1 Institute of Infection, Immunity and Inflammation College of Medical, Veterinary and Life Sciences University of Glasgow, Glasgow, 2 Centre de Résonance Magnétique des Systèmes Biologiques, Université Bordeaux Segalen, Bordeaux, France

Transketolase is an enzyme of the non-oxidative branch of the pentose phosphate pathway. The enzyme has been shown to be localised to both the glycosomes and cytosol in *Leishmania mexicana*. Here we show that localisation is regulated by the C-terminus of the protein and the relative distribution to the glycosomes or cytosol can be regulated by altering the length of the C-terminal tail of the enzyme. By manipulating the sub-cellular localisation we could determine the enzyme's contribution to metabolism in the respective compartments. When deprived of transketolase, through gene knockout, promastigote cells adopt a "stringent" metabolism, consuming two times less glucose and reducing the secretion of end products. No apparent increase in amino acid catabolism accompanied the loss of glucose use. Transketolase deficient cells have reduced ability to infect macrophages and they did not survive as axenic amastigote forms. Interestingly, the ability of *L. mexicana* to infect macrophages is also depended on the sub-cellular localisation of transketolase.

Immunization trials with native and recombinant proteins of *Fasciola gigantica* in buffalo (P199)

Ravikumar G1, Raman M1, Asha Alex1, Julie Martina1, Satish L2, Balaji S3, Suganya Devi P1, Raina OK3
1 Zoonoses Research Laboratory,Centre for Animal Health Studies, Tamilnadu Veterinary and Animal Sciences University, Chennai, 2 Department of Veterinary Parasitology, Madras Veterinary College, 3 Indian Veterinary Research Institute, India

Fasciola gigantica is an economically significant pathogen in India. The disease is reported throughout the country. The disease results in loss of production, poorer milk- fat percentage and can impair immune functions to alter diagnostic testing to other diseases. This is also classified as a zoonotic disease by the WHO. While resistance to anthelminthics is on the rise, and with no new anthelminthic licensed in the past three decades the search is for the exploitation of certain proteins of the parasites as immunogens in the form of a prototype vaccine. Native ES antigens and recombinant sigma Glutathione transferase were the immunogens used with oil emulsion adjuvant approved for use in food animals. Humoral and cell mediated immunity was studied using an ELISA test and cell proliferation using MTT dye reduction test. The animals were challenged with metacercariae. The results showed that the immunogens with adjuvant were clinically safe, and significant humoral and cell mediated pattern in the test as against the control group. This trial was done with one dose of antigen; further work is required with varied doses for a small scale field study in subsistence and organized farm conditions.

An alternative model for the role of RP2 in flagellum assembly in the African trypanosome (P200)

Paul G. McKean Jane Andre, Louise Kerry, Xin Qi, Erica Hawkins, Kristina Drižytė, Michael L. Ginger
Faculty of Health and Medicine, Biomedical and Life Sciences, Lancaster University, Lancaster, LA14YQ, UK

TbRP2 is a basal body located tubulin cofactor C (TBCC) domain-containing protein essential for axoneme formation in *Trypanosoma brucei.* We previously reported that RNAi mediated ablation of TbRP2 leads to the loss of intense labelling of the trypanosome basal body by the monoclonal YL1/2 (classically used to detect tyrosinated alfa-tubulin). This led us to previously suggest TbRP2 plays a critical role in the recruitment/processing of tubulin heterodimers destined for axonemal incorporation; thus explaining the flagellum assembly defects that arise from loss of TbRP2 expression. However, there is debate as to whether the flagellar assembly function of specialised, basal body TBCC-domain containing proteins such as RP2 is associated with tubulin processing, or whether RP2 serves a more general vesicular trafficking function in flagellum assembly (as proposed for the human RP2 ortholog). In this study we now reveal; (i) over-expression of a C-terminal myc-tagged TbRP2 variant also results in loss of YL1/2 labelling - but has no effect on assembly of the trypanosome flagellum, (ii) TbRP2 itself encodes a YL1/2 epitope, and (iii) the flagellum biogenesis defect seen in TbRP2 RNAi mutants could also be explained as a function of failing to recruit evolutionary conserved ciliary gate proteins (the ciliary gate is located at the base of the flagellum and influences flagellum protein content). Taken together our new data indicates the role of TbRP2 in trypanosome flagellum assembly is undoubtedly more complex than our previous (tubulin processing) model suggested, and reveals hitherto unappreciated association between RP2 and cilary gate components in flagellated eukaryotes.

Using Molecular Approaches to Identify Cidal or Static Activity of Antimalarials (P201)

Ane Rodriguez Alejandre, Noemí Bahamontes-Rosa, Rubén González-del Río, María G. Gómez-Lorenzo and Francisco Javier Gamo-Benito
GSK, Madrid Tres Cantos

Malaria is still a life threatening disease, responsible for a million deaths worldwide every year, although it is preventable and treatable. Because of the continuous spread of resistance to current antimalarial treatments, there is an urgent need of developing new drugs to fight against the malaria parasites responsible for the illness. In 2010, a project carried out at GlaxoSmithKline identified, using a phenotypic high throughput screening, 13,533 potent chemical inhibitors of the intraerythrocytic growth of *P. falciparum*. However, new biological tools are needed to identify, in a fast way, the most competitive compounds to enable new antimalarial treatments. In a previous work (Bahamontes-Rosa et al, 2010), it was presented a fast, reliable and reproducible molecular method that discriminates between cidal and static behavior of antimalarials. The method is based on the quantification of mRNA levels after drug treatment. Here, we present the results using compounds selected from different lead optimization programs at GSK. As a noticeable finding, this methodology is able to detect induction of gametocytogenesis due to the action of some drugs. This assay can facilitate the discovery of new efficacious antimalarials in a fast and efficient manner.

Insight into the evolution of nuclear envelope proteins from diverse Eukaryotes (P202)

Ludek Koreny1, Jennifer M. Holden1, Samson Obado2, Michael P. Rout2,Mark C. Field1,
1 Division of Biological Chemistry and Drug Discovery, University of Dundee, Dundee, Scotland, 2 The Rockefeller University, New York, USA

The nuclear envelope (NE) is the defining feature of the eukaryotic cell but until recently, most of the characterized NE proteins were only known from Opisthokonts, the eukaryotic supergroup that includes fungi and animals. These proteins include components of the nuclear pore complex (NPC), nu-clear lamina and SUN/KASH domain proteins, which serve as bridges con-necting nucleoskeleton with the cytoskeloton. Here we discuss the distribution of NE components based on pan-eukaryotic homology searches and phylogenetic analyses together with recent experimental data on the composition of the NE from the diverged eukaryotic parasite *Trypanosma brucei*, to understand in more detail the evolution of these complex cellular structures. We find that an NPC very similar to that in humans was already pre-sent in LECA. Some of the proteins that fail to be identified by homology searches in diverged eukaryotes may be present as exemplified by proteomic studies on *Trypanosoma*, where many nucleoporins with typical secondary structures were found despite the lack of sequence conversation. By contrast, several components evolved after the diversification of eukaryotes. The trypanosome nuclear basket is composed of distinct proteins compared with opisthokonts but its functions are very similar and may thus serve as an example of convergent evolution. This may also be the case for the NUP-1, a trypanosome specific protein with roles similar to lamins. The divergence of this group of protists is further underlined by the apparent absence of the classical SUN/KASH domain protein complexes, which are otherwise conserved among eukaryotes and were clearly present in LECA.

Filling in the gaps in helminth diversity: preliminary comparative analyses of 60 draft genomes for parasitic worms (P203)

James A. Cotton1, Isheng Jason Tsai1, Eleanor Stanley1, Bhavana Harsha1, Avril Coghlan1, John Martin2, Phillip Ozersky2, Kym Pepin2, Xu Zhang2, Bruce Rosa2, Rahul Tyagi2, Xin Gao2, Daria Gordon1, The Helminth Genome Initiative Consortium, Nancy Holroyd1, Makedonka Mitreva2, Matthew Berriman1
1 Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge. CB10 1SA, UK, 2 The Genome Institute at Washington University, Washington University School of Medicine, Saint Louis, Missouri, USA.

Genomic data is available for a growing number of helminth species, including exemplars of most major clades of parasitic worms, but these data do not capture much of the diversity of helminth parasites. To address this, the helminth genome initiative at the Sanger institute is attempting to expand our knowledge of helminth genomes. With a large group of collaborators, we have generated whole-genome shotgun sequence data and draft genome assemblies for over 50 parasitic nematode and platyhelminth species for which genomic data was previously unavailable, including parasites of medical and veterinary importance and some comparative species that occupy important phylogenetic positions, for example as outgroups to important groups of parasites. These data include the first genomic data for a number of important parasitic groups. The genomes we include are demonstrably largely complete, but the fragmentary nature of the assemblies makes some analyses challenging. Despite this, we can see for the first time the broad outlines of genome evolution for parasitic helminths, and take a phylostratigraphy approach to assess how well *C. elegans*, and the better-known parasite species in each group represent the diversity of helminths.

Molecular diversity of sperm proteins provides insights into the speciation and the lack of genetic barriers between species of African Schistosoma blood flukes (P204)*

Toby L. Landeryou1, Juliet P. Dukes1, Aidan M. Emery2, David Rollinson2, Scott P. Lawton1
1. Kingston University, London; 2. Natural History Museum, London

Throughout Africa there are several Schistosoma blood fluke species, several of which are known to be sympatric not only in geographical range but also sharing the same snail intermediate hosts and definitive host species. Many species of Schistosoma can interbreed with each other and produce viable young with several hosts (both human and animal) being shown to be infected with hybrid offspring. This indicates a breakdown of speciation boundaries and illustrates a lack of genetic barriers that would prevent successful hybridisation. Genes coding for sperm proteins associated with fertilisation of the egg and sperm flagella locomotion diverge rapidly in most invertebrate species, creating important species barriers. In other organisms such as marine snails and rotifers that readily hybridise there has been a distinct lack of divergence within sperm associated proteins allowing the formation of viable zygotes. In this study, three sperm protein associated genes; beta 1,4 galactotransferase, sperm nuclear basic protein and antigenic sperm protein 1. These

were identified using the *S. mansoni* genome resource and then sequenced across several closely related species from both the *S. mansoni* and *S. haematobium* groups. Low levels of genetic diversity were observed between species within the groups but large amounts of variation occurred between the *S. mansoni* and *S. haematobium* groups. A high level of nonsynonymous mutations was recorded leading to marked differences in the properties of the sperm proteins. These results indicate a recent, rapid divergence of the Schistosoma species groups but does question the validity of some of the species

Allergenic proteins are targets for mammalian IgE mediated immune response against metazoan parasites (P205)*

Nidhi Tyagi1, Edward Farnell2, Colin Fitzsimmons2, Stephanie Ryan3, Rick Maizels3, David Dunne2, Janet Thornton1, Nicholas Furnham1,4

1 The EMBL-European Bioinformatics Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SD, United Kingdom, 2 Department of Pathology, University of Cambridge, Tennis Court Road, Cambridge CB2 1QP, United Kingdom, 3 Institute of Immunology and Infection Research, University of Edinburgh, United Kingdom; 4 London School of Hygiene and Tropical Medicine, Keppel Street, London, WC1E 7HT, United Kingdom

Allergenicity can be described as an uncontrolled and hostile Type 1 hypersensitive immune reaction towards environmental antigens from diverse sources such as foods, plants and various organisms. These reactions are mediated by T helper cell type 2 (Th2) response through IgE production, which were originally evolved to provide immunity in mammals against metazoan parasites such as helminths and arthropods. Similar IgE mediated immune responses in allergenic conditions (unregulated) and in acquiring immunity against pathogens (regulated) can be attributed to a coincidental molecular similarity between antigens and pathogenic proteins. The current study focuses on detecting IgE inducing structure motifs in proteins encoded in genomes of parasites that share molecular similarity with epitopic regions of allergens. 2385 experimentally verified epitopic regions from 205 allergenic proteins represented in highly populated protein domain families have been culled from IEDB database and searched in proteomes of *Ascaris lumbricoides, Brugia malayi, Schistosoma mansoni, Echinococcus granulosus, Onchocera volvulus, Taenia* sp. and *Trichuris trichiura* for recognition of epitope-like regions in parasitic proteins by employing various computational approaches. Comprehensive analysis of these significantly similar epitopic and epitope-like regions were performed by comparing their topology on 3D structures/models of allergenic and parasitic proteins respectively. Detailed sequence and structure based analyses highlight that epitopic regions from allergens share common molecular features with parasitic proteins and thus support the hypothesis that allergens are targets for mammalian IgE mediated immune responses. Findings from this study further enrich our understanding of allergenicity in light of immunity.

The whipworm *Trichuris muris*: dual-species transcriptomics of an intimate host-pathogen interaction (P206)

Adam J. Reid1, Bernardo J. Foth1, Isheng J. Tsai1,2, Allison J. Bancroft3, Sarah Nichol1, Alan Tracey1, Nancy Holroyd1, James A. Cotton1, Eleanor J. Stanley1, Magdalena Zarowiecki1, Jimmy Z. Liu4, Thomas Huckvale1, Philip J. Cooper5,6, Richard K. Grencis3, Matthew Berriman1

1 Parasite Genomics, Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SA, UK, 2 Division of Parasitology, Department of Infectious Disease, Faculty of Medicine, University of Miyazaki, Miyazaki 889-1692, Japan, 3 Faculty of Life Sciences, University of Manchester, Oxford Road, Manchester M13 9PT, UK, 4 Statistical Genetics, Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge CB10 1SA, UK, 5 Liverpool School of Tropical Medicine, Pembroke Place, Liverpool L3 5QA, UK, 6 Centro de Investigación en Enfermedades Infecciosas, Escuela de Biología,Pontificia Universidad Católica del Ecuador, Quito, Ecuador

Whipworms are common soil-transmitted helminths that cause debilitating chronic infections in man. These nematodes are only distantly related to *Caenorhabditis elegans* and have evolved to occupy an unusual niche, tunneling through epithelial cells of the large intestine. We have produced a high quality genome assembly of the murine laboratory model *Trichuris muris*. We have used this as a basis for transcriptome sequencing to identify genes in host and parasite, which are involved in host-parasite interaction. The anterior part of the worm tunnels into the intestine and contains the specialized bacillary band and stichosome tissues, found only in whipworms and related parasites. We compared the transcriptome of this region with that of the largely sexual posterior and identified two gene families that we hypothesise are involved in interaction with the host immune system. We went on to generate RNA-seq data from chronically whipworm-infected mouse caecum and used this to explore a caecum-wide regulated Th1-like immune response in unprecedented detail. We also used our results to explore the similarities between chronic whipworm infection and immunity-related diseases such as ulcerative colitis.

Filarial genome annotation in WormBase (P207)

Eleanor Stanley, James Cotton, Nancy Holroyd, Sara Lustigman, Michael Paulini, Kevin Howe, Matt Berriman
Wellcome Trust Sanger Institute, Hinxton, Cambridge, CB10 1SA, UK The EMBL-European Bioinformatics Institute, Wellcome Trust Genome Campus, Hinxton, Cambridge, CB10 1SD, UK Laboratory of Molecular Parasitology, Lindsley F. Kimball Research Institute, New York Blood Center, 310 E 67th St, New York, NY 10065, USA

WormBase is an international consortium of biologists and computer scientists dedicated to providing accurate, complete, accessible information concerning the genetics, genomics and biology of the *Caenorhabditis elegans* and selected nematodes of medical, and agricultural importance. These include a number of human, animal, plant and insect parasites. Extending a model organism database to cope with the diversity of parasitic nematodes presents several key challenges, not least is the small size of each species' research community. WormBase have traditionally undertaken manual curation of gene models for *C. elegans* and other *Caenorhabditis* species. In 2011, this remit was expanded to include selected parasitic nematodes, beginning with *Brugia malayi*. An intensive program of manual curation resulted in the review/revision of nearly one third of the genes suggested by the Brugia community. A major new addition, available in WormBase release 241, is the genome of the human filarial parasite species, *Onchocerca volvulus*, which causes Onchocerciasis or "River Blindness", a neglected tropical disease that affects over 37 million people, mainly in Africa. A single adult worm of *Onchocerca volvulus* from Republic of Cameroon has been used to produce a reference genome by deep sequencing and optical mapping. The resulting chromosome-sized scaffolds have been annotated using Maker. To enable the research community to better contribute to this important genome project, the gene model annotations are now available in WormBase. As with Brugia, WormBase has begun manual curation of gene models, and we welcome feedback from the Onchocerca community in order to help identify and prioritise curation targets.

Epidemiological studies on co-infection with tuberculosis and intestinal parasites in Benue State, Nigeria (P208)

Edward Omunu, Idu, M. E., Amuta, E. U.
Benue State, Nigeria, West Africa Department of Biological Sciences Benue State University Makurdi Nigeria.

This study investigated 2,519 participants for co-infection with *Mycobacterium tuberculosis* and intestinal parasites in some communities in Benue State, Nigeria. Tuberculosis diagnosis was done using the Ziehl-Neelseen's staining technique to identify acid fast bacilli (AFB). The formalin-ether concentration technique was used to detect ova of intestinal parasites. The prevalence of intestinal parasites in participant referred for TB test in health facilities was 41.9%. For participants who were already positive for TB, the prevalence was 38.4% and for apparently healthy participants it was 40.3% ($\chi 2 = 157$, df = 2, $P > 0.05$). The overall prevalence of *M. tuberculosis* was 17.6%, though the prevalence of 30.1% was recorded in participants who were already on the DOTS treatment programme. The sputum smear positive cases was significant in all LGAs investigated ($\chi 2 = 387$, df = 6, $P < 0.05$). The prevalence of intestinal parasites in participants with severe TB (51.2%) was significantly higher than those who had scanty infection (43.6%) and those who were negative (38.6%) ($\chi 2 = 173$, df = 3, $P < 0.05$). The odds of being infected with intestinal parasites increased with severity of TB in the study area. Co-infection with *M. tuberculosis* and intestinal parasites was significantly higher in male participants ($P < 0.05$). Prevalence of polyparasitism was higher in participants with severe TB and *Entamoeba histolytica* and hookworm were the most prevalent parasite combination encountered.

***In vitro* characterization of a new chemotype active against asexual and sexual *Plasmodium falciparum* parasites (P209)**

Silvia Parapini1, Andrea Pancotti2, Nicoletta Basilico3, Yolanda Corbett1, Sarah D'Alessandro1, Donatella Taramelli1, Sergio Romeo2
1 Dipartimento di Scienze Farmacologiche e Biomolecolari, University of Milan, Milan Italy, 2 Dipartimento di Scienze Farmaceutiche, University of Milan, Milan Italy, 3 Dipartimento di Scienze Biomediche, Chirurgiche ed Odontoiatriche, University of Milan, Milan Italy.

The malaria control measures recommended by WHO include drug treatment with artemisinin-based combination therapy (ACT). However, cases of clinical resistance to ACT have been reported in SE-Asia. To substitute artemisinin derivatives and to achieve the goals of WHO malaria elimination/eradication agenda, new antimalarials able to kill the transmissible sexual stages of the parasite or to prevent the parasite development in the mosquito are strongly needed. We recently discovered a new chemotype with promising characteristics. The structural core is constituted by a 4,4'-oxybisbenzoic acid linker bound to two amino acids. Derivative DC18 has been selected as a novel hit, because as

molecule offers several advantages: good water-solubility, Lipinski's rules of five are fulfilled except for the molecular weight (MW=579), fast, affordable and low cost synthesis. DC18 possess a very high activity *in vitro* (0.5-5nM) against different *Plasmodium falciparum* strains (CQ-S and CQ-R) including fresh clinical isolates. Preliminary data show that DC18 reacts very fast, mostly killing ring stage parasites, but also trophozoites. In addition, DC18 is active on both young (IC50 150nM on stage II-III) and mature Pf gametocyte (IC50 250nM on stage IV-V). DC 18 is not toxic against human endothelial cell (IC50 >20 µM) and also showed additive properties with chloroquine. The molecular target is presently under study applying a selective and sensitive chemical proteomic approach. The similarity with artemisinin (high activity, fast time to kill, effects on rings stages), makes DC18 a very interesting compound for future development for malaria combination therapy.

Red Queen Communities: Antagonistic coevolution in multi-species communities (P210)*

Suzanne Ford, Kayla King
Department of Zoology, University of Oxford, The Tinbergen Building, South Parks Road, Oxford OX1 3PS, UK

The Red Queen hypothesis states that an agent in a coevolutionary interaction needs to continually adapt in order to maintain its fitness relative to its opponent. The hypothesis is named after a passage in Lewis Carroll's 'Through the looking glass' where the Red Queen tells Alice 'in this place it takes all the running you can do to keep in the same place'. In the study of host-parasite coevolution, this hypothesis is traditionally considered between a single host species and a single parasite species. In nature, however, where hosts are home to a whole microbial community, such an interaction rarely exists in isolation. Consequently, the impact of co-inhabiting microbes on host-parasite coevolutionary dynamics is likely to be of major effect and is therefore considerably understudied. In addition to this, little is still known about the effect of co-inhabiting microbial species on the evolution of parasite virulence, a key component of disease systems. As such, I aim to investigate the effect of multi-microbial communities on the dynamics of host-parasite coevolution and parasite virulence evolution, including the role of coevolutionary arms races between microbes within hosts and the role of direct versus indirect microbial interactions. These studies will use a *Caenorhabditis elegans* (host)–bacteria (microbe) system in an experimental evolution framework and will exploit time-shift analyses. Initially, I am coevolving two parasite species, *Staphylococcus aureus* and *Enterococcus faecalis* within a non-evolving *C. elegans* host in order to investigate the dynamics of parasite-parasite arms races within hosts and the effect on parasite virulence.

***Blastocystis* sp. in wild life (water monitor lizard, mouse-deer and Malayan porcupine) of Tioman Island, Malaysia (P211)**

Mohd Zain S.N., Woh, P. Y., Farah Haziqah M.T. , Suresh Kumar, Govind
Institute of Biological Sciences, Faculty of Sciences, University of Malaya, 50603 Kuala Lumpur, Malaysia. Department of Parasitology, Faculty of Medicine, University of Malaya, 50603 Kuala Lumpur, Malaysia.

Numerous reports of *Blastocystis* sp. infection have been conducted in animals including, domestic pets, live stocks and in particular, in non-human primate species. However, none have been made to study the presence of *Blastocystis* in wildlife in Malaysia. The present study is the first ever report on the presence of *Blastocystis* infection from three wildlife species on Tioman Island, which consisted of six water monitor lizard (*Varanus salvator*), four mouse-deer (*Tragulus* sp.) and one Malayan porcupine (*Hystrix brachyura*). The protozoan was screened using light microscopy and cultured *in vitro* in Jones medium. *Blastocystis* species was detected from one of the six water monitor lizards and a mouse-deer. The vacuolar form was the most common cell form found in cultures similar to the human isolate, *B. hominis*. The optimal culture temperature for the reptilian isolate was lower at 24ºC but higher for the isolate from the mouse-deer at 37˚C. The *Blastocystis* isolate from the monitor lizard showed consistency in growth and was successfully maintained in the laboratory using Jones medium. Unfortunately, the mouse-deer isolate failed to survive.

Genetic variation in recently-wild genotypes of the parasitic nematode *Strongyloides ratti* (P212)

Diogo Ribeiro, Avril Coghlan, Victoria Hunt, Nancy Holroyd, Mark Viney, Matthew Berriman
Wellcome Trust Sanger Institute and University of Bristol.

Parasitic nematodes are significant animal pathogens, with a quarter of the human population, predominantly in the developing world, infected with nematodes. Strongyloides is a genus of about 50 species of obligate gastrointestinal nematode parasites, which infect mammals, birds, reptiles and amphibians. *S. stercoralis* is a common parasite of humans infecting some 100-200 million persons worldwide and host immunosuppression can lead to disseminated

strongyloidiasis, which is fatal if not treated. *S. ratti* is a common rat parasite and is the laboratory analogue of the human parasite *S. stercoralis*. *S. ratti* has been extensively studied and its genome has been sequenced. To investigate genomic diversity in S. ratti we have analyzed the patterns of variation (e.g. SNPs) in 10 different isofemale lines derived from 10 wild isolates collected around the world. Here we present an analysis of differences in SNP density along *S. ratti* chromosomes, and highlight interesting functional classes of genes that show unusually high diversity. Phenotypic data from these *S. ratti* lines show differences in some infection characteristics, such as fecundity and reaction to the host's immune response. We plan to explore the dataset of *S. ratti* genomic diversity to investigate which genetic diversity underlies these phenotypic differences.

A novel target for the treatment of Nematode infections (P213)*

George Cherian Pandarakalam, B. Connolly, B. Mueller, J. Pettitt, L. Philippe
Institute of Medical Science, University of Aberdeen

There are more than one million species of nematodes and most of them are free-living but still there are tens and thousands of parasitic nematodes. These parasitic nematodes cause a variety of infections in both humans and animals and are also responsible for huge economic losses caused by yearly crop damage. Repeated usage of anthelmintics against these parasitic nematodes has resulted in the development of resistance among nematodes. So there is an urgent requirement for the development of new anthelmintics that target novel sites. An ideal target should be present only in the nematode and not in the host vertebrates and plants. And such a promising target could be Spliced leader (SL) *trans*-splicing. SL *trans*-splicing is a process that occurs during mRNA maturation in certain groups of invertebrates like nematodes and involves the addition of a short (20-40nt) RNA, derived from a non-polyadenylated RNA called the spliced leader RNA, to the 5' end of a subset of pre-mRNAs. SL *trans*-splicing has been identified in all nematode species studied to date, suggesting that it is a phylum-wide process, but it is absent in vertebrates and plants, the major hosts for parasitic nematodes. Hence, SL *trans*-splicing is a potential target for novel anthelmintics. Using a GFP based *in vivo* assay developed in the model organism *Caenorhabditis elegans*, our aim is to identify novel compounds that will specifically inhibit SL *trans*-splicing in nematodes and to explore the potential to develop those compounds into patentable drugs.

Snapshot profiling of the activity of current anti-leishmanial drug candidates against intracellular *Leishmania* in a panel of host cells (P214)

Markella Koniordou1, Susan Wyllie2, Karin Seifert1
1 London School of Hygiene & Tropical Medicine, Faculty of Infectious and Tropical Diseases, Keppel Street, London WC1E 7HT, 2 Division of Biological Chemistry and Drug Discovery, Wellcome Trust Biocentre, University of Dundee, Dundee, Scotland, DD1 5EH

The leishmaniasis are a spectrum of vector-borne diseases caused by obligate intracellular parasites of the genus *Leishmania*. There is no efficient vaccine available and current treatment options are far from satisfactory. Within the human host *Leishmania* parasites survive and multiply as intracellular amastigotes in the parasitophorous vacuole of macrophages and biological evaluations rely on *in vitro* models that simulate the physiological environment of the *Leishmania* parasite in the mammalian host. Thus the intracellular amastigote stage is the clinically relevant and often used life cycle stage of *Leishmania* in *in vitro* anti-leishmanial drug activity evaluations. However different host cells have been used in the research community and we previously demonstrated differences in anti-leishmanial drug activity against intracellular *L. donovani* in different host cells. In the work presented here we have 1) expanded the range of drugs investigated for host cell dependent drug action to include current anti-leishmanial drug candidates from different chemical classes and 2) characterised different commonly used host cells with attention to key factors in drug action

Evaluation of Prostaglandin F2-alpha synthase expression by *Leishmania braziliensis* (P215)

Eliza V.C. A. Ferreira, Tiago R. Ferreira 1, Paul Kaye2, Angela K. Cruz1
1 Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, Brazil, 2 Centre for Immunology and Infection, University of York, York, UK.

Prostaglandin (PG) production is not restricted to mammals; recent studies have shown parasites able to synthesize PG from metabolites of arachidonic acid (AA). Trypanosomes carry Prostaglandin F2a synthase (PGF2S) enzyme and produce prostaglandin F2alpha (PGF2a). The PGF2S of *Leishmania braziliensis* (LbrPGF2S) shares 34% identity and 51.4% similarity with the mammalian homolog (AKR1C3). LbrPGF2S has been detected in the secretome of L. braziliensis and in the exosome of *L. donovani*. Also, according to the TDR Targets Database the *L. major* PGF2S homolog has 13 putative

antigenic epitopes, with 77.8% antigenicity and druggability index of 0.8 (range:0-1), making it one of the most antigenic proteins of *L. major* and a putative candidate as drug target. In this work, we have generated LbrPGF2S overexpressor, which was confirmed by Southern and Western blotting. The level of intracellular LbrPGF2S expression was evaluated in the presence or absence of AA, at 26°C or 37°C. Parasites cultivated for 3 days and 7 days with AA have shown a decreased LbrPGF2S expression. Although the LbrPGF2S overexpressor and its control showed no differences in axenic growth curve, the *in vitro* infection profile displayed a positive correlation between increased levels of LbrPGF2S expression and parasite survival within the host cells. The analyses of LbrPGF2S secretion and PGF2a production in axenic culture and in infected host cell culture are underway. Our preliminary results emphasize the importance of understanding how LbrPGF2S affects the host infection.

High Throughput Intramacrophage (INMAC) assay for screening compounds against *Leishmania donovani* (P216)*

Sujatha Manthri, Manthri S, De Rycker M, Gray DW.
Drug Discovery Unit, James Black Centre, University of Dundee, DD1 5EH

Leishmania donovani is the causative agent of visceral leishmaniasis, a neglected tropical disease with an estimated 300,000 cases and 20,000 deaths annually. There is an urgent need for new treatment therapies as the current treatment options involve high cost, lengthy treatment regimes, toxicity and emerging drug resistance. *Leishmania donovani* leads a digentic life -cycle with the free living promastigote stage developing in the gut of the sandfly and the mammalian amastigote stage dividing and multiplying in the parasitophorous vacuole of the macrophage.Several anti-leishmanial screening assays for discovering new drugs described in literature involve the free living forms of the parasite due to the ease of maintaining them in a laboratory environment and ease of screening large volume of compounds. However this is not a true representation of the environment in which the parasite survives. Here we describe a high throughput, high content image based intramacrophage (INMAC) assay for screening compounds against *Leishmania donovani*. The method utilizes 384 well plates and uses the THP-1 (human monocytic leukemia) cell line and eGFP expressing *Leishmania donovani* (MHOM/SD/62/1S-CL2D,LdBOB) amastigote stage. This method has allowed us the flexibility of primary screening and follow up by potency screening of interesting molecules.

The genome sequence of the Cassava parasite *Phytomonas francai* provides insight into the evolution of parasitism in kinetoplastids (P217)

Claire E. Butler1, David Emms1, Richard Wheeler2, Steve Kelly1
1 Department of Plant Sciences, University of Oxford, Oxford UK, 2 Sir William Dunn School of Pathology, University of Oxford, Oxford, UK.

Phytomonas francai is a kinetoplastid from the order Trypanosomatida which parasitizes the cassava plant, *Manihot esculenta*. The parasite is transmitted by an insect vector directly to the latex vessels of the plant and infection has been linked to the disease empty root syndrome which can devastate whole harvests. Within the insect vector the parasite is present in a promastigote-like form which can be cultured axenically, these forms undergo cell division in a mode similar to *Leishmania mexicana* with uneven elongation of the flagellum. Here we sequenced the genome of *P. francai* and use it (with other available *Phytomonas* resources) to perform an analysis of genome evolution analysis across all sequenced trypanosomatids. We developed a novel orthologue identification method coupled with evolutionary inference and ancestral genome reconstruction to predict the gene family gains and losses that have occurred in kinetoplastid evolution. Furthermore we use this novel reconstruction approach to identify putative pathogenicity islands in the genome of *Leishmania mexicana*. This study provides extensive novel insight into the evolution of parasitism in kineplastids and provides a new comparative genomic approach to identify parasite factors important for host specificity.

Ordering components of the slender to stumpy signalling pathway in *Trypanosoma brucei* (P218)*

Lindsay McDonald , Keith R. Matthews
Institute for Immunology and Infection Research, School of Biological Sciences, University of Edinburgh, Edinburgh EH9 3JT, UK

In the mammalian bloodstream, *Trypanosoma brucei* undergo differentiation from proliferative slender forms to arrested, transmissible, stumpy forms. This transition is associated with extensive morphological and metabolic changes that enable adaptation to the contrasting environment of the tsetse vector midgut but also influences infection dynamics within the mammalian host. However, the molecular pathways mediating this transformation remain uncharacterised.

Recently, a number of candidate genes involved in this process were identified using an RNAi library screen, in which cells were selected for resistance to pCPTcAMP, a membrane-permeable cyclic AMP analogue that induces differentiation. One candidate, MEKK1 (MAPK/ERK kinase kinase 1), belongs to the MAPK family of signalling proteins. RNAi silencing of MEKK1 in a differentiation-competent pleomorphic strain results in decreased responsiveness to pCPTcAMP *in vitro* and decreased differentiation to stumpy form in mice. The unusual kinetoplastid-specific target of rapamycin kinase TOR4 has previously been shown to act as a negative regulator of stumpy formation in monomorphs. RNAi of TOR4 in a pleomorphic strain similarly increased differentiation, an effect antagonistic to that of MEKK1 and other stumpy formation candidate genes. Therefore, simultaneous RNAi of the stumpy inhibitor TOR4 and genes that promote stumpy formation should enable ordering of different determinants of differentiation relative to each other, enabling the pathways controlling this event to be delineated. Simultaneous RNAi of TOR4 and a NEK kinase, one such candidate, resulted in reduced growth and intact responsiveness to pCPTcAMP, indicating that loss of TOR4 is dominant and is therefore likely to act downstream or independently of NEK.

Invasion of the tsetse midgut by *Trypanosoma brucei*: a role for the metalloprotease MSP-B? (P219)

Naomi A. Dyer1, Samirah Perally1, Clair Rose1, Michael J. Lehane1, Alison J. Beckett2, Ian Prior2, Alvaro Acosta-Serrano1
1 Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA, 2 Department of Cellular and Molecular Physiology, Crown Street, University of Liverpool, L69 3BX

Trypanosoma brucei breaches the peritrophic matrix (PM) of the tsetse fly to colonize the extraperitrophic space of the midgut. Although this process appears essential for trypanosomes to be transmitted by their tsetse hosts, the molecular mechanism(s) by which breaching of the PM occurs is not well understood. Unlike *Plasmodium* and *Leishmania*, which use chitinases to cross the PM of their respective invertebrate hosts, *T. brucei* lacks chitinase activity. This suggests that other parasite enzymes may be involved in degrading the tsetse PM. Here we present initial steps to test a possible alternative mechanism of PM invasion by the *T. brucei* metalloprotease MSP-B. MSP-B is a GPI-anchored MSP involved in the removal of the VSG coat during the stumpy to procyclic transition, but remains expressed in midgut forms. We have generated RNAi procyclic clones, which are currently being tested for their ability to colonize the tsetse gut when the expression of MSP-B is down-regulated. In addition, we will present ultrastructural data showing the damage caused to the tsetse PM during *T. brucei* infection.

In vitro* and *in vivo* analysis of pyrimidine requirements in bloodstream forms of *Trypanosoma brucei* (P220)

Maria Valente, Daniel García-Caballero, Víctor Castillo-Acosta, Antonio E. Vidal, Luis M. Ruiz-Pérez, Dolores González-Pacanowska.
Instituto de Parasitología y Biomedicina "López-Neyra". CSIC. Parque Tecnológico de Ciencias de la Salud, Granada, Spain.

A balanced pool of deoxyribonucleotides is essential for DNA replication and repair and disturbances in the supply of dNTPs may lead to genetic mutations and cell death. Deoxyribonucleotides can be supplied in most living organisms by two pathways, the de novo and the salvage pathway. *Trypanosoma brucei* and in general kinetoplastid parasites have both pathways which suggests that they can compensate for each other and that neither processes are essential although *in vivo* studies are required to establish the true requirements in a physiological context. Indeed de novo pyrimidine synthesis up to UMP is dispensable and cells are capable of surviving in the presence of uracil both *in vitro* and *in vivo*. However, while UMP synthesis is not essential, dTMP formation *in vivo* is dependent on a unique bifunctional dihydrofolate reductase-thymidylate synthase that uses dUMP as substrate. Deoxyuridine triphosphate nucleotidohydrolase (dUTPase) converts dUTP/dUDP into dUMP, the unique precursor for synthesis of dTMP, and maintains at the same time a low dUTP/dTTP ratio, thus preventing the incorporation of dUTP during replication. We have analysed the pyrimidine requirements of dUTPase null mutants. Surprisingly deoxyuridine cannot reverse the lethal phenotype and out of the several nucleosides and nucleobases tested, only thymidine and 5-methyl deoxycytidine are capable of supporting parasite growth *in vitro.* We also show that dUTPase null mutants cannot establish an infection *in vivo*. These observations indicate that (i) only thymidine salvage can efficiently modulate the dUTP/dTTP ratio (ii) the existence of an active cytidine deaminase involved in pyrimidine nucleoside interconversion.

The role of Variant Surface Glycoprotein in *Trypanosoma brucei* evasion of phagocytosis by macrophages (P221)*

Jackie Lok-Yee Cheung, Nadina Vasileva Wand, Gloria Rudenko
Department of Life Sciences, Imperial College London, London, UK, SW72AZ

The Variant Surface Glycoprotein (VSG) coat of bloodstream form (BSF) *Trypanosoma brucei* is essential for evasion of the host immune system. During infection, VSG-specific antibodies bind to the VSG coat but BSF *T. brucei* can clear low titres of surface bound antibodies effectively through high rates of VSG recycling. Blocking the synthesis of VSG using tetracycline inducible RNAi triggers a precytokinesis cell cycle arrest and a global block in translation. In mouse infections, trypanosomes are cleared rapidly when VSG RNAi is induced, however, the mechanism for this clearance is poorly understood. In this study, we investigated the role of macrophages in this clearance using immunofluorescence microscopy of trypanosomes co-cultured with macrophages. In the presence of a VSG-specific antibody but not complement, we observed a 2.6-fold increase of phagocytosis of trypanosomes where VSG synthesis had been blocked. We also show that blocking VSG synthesis results in reduced anti-VSG antibody clearance on the trypanosome cell surface, suggesting impaired recycling of VSG-antibody complexes. Our data suggest that antibody-VSG complexes are key in the recognition and phagocytosis of trypanosomes by macrophages.

Assessment of the *Trypanosoma rangeli* proteome (P222)

Edmundo C. Grisard1, Débora D. Lückemeyer1, Glauber Wagner2, Patricia H. Stoco1, Hercules Moura3, John Barr3, Mário Steindel1,

1 Universidade Federal de Santa Catarina, Florianópolis, SC, Brazil, 88040-970, Brazil, 2 Laboratório de Doenças Infecciosas e Parasitárias, ACBS/Unoesc, Brazil, 3 Flu and Toxin Laboratory, CCB/DLS/NCEH/CDC, USA,

Trypanosoma rangeli is a non-pathogenic parasite of Latin American mammals, for which, limited proteomic information is available. Soluble proteins obtained from parasites forms during the differentiation process were analysed using gel-based (1-D and 2-D) and gel-free (LC-MS/MS) proteomic approaches. For 1-D, proteins were resolved in 4-12% Nu-Page SDS-PAGE and for 2-D profiles, proteins were resolved using pH 3-10 strips (13 cm) and then in 12% SDS-PAGE. A total of 375 gel slices and 1,057 spots were submitted to in-gel digestion. For the gel-free analysis, 70 µg of soluble proteins were digested by RapGest treatment. Tryptic peptides were analysed by LC-ESI-MS/MS, identifying 1,455 non-redundant T. rangeli proteins (FDR <0.4%) (Gel-free=716, 1-D=1,410, 2-D=182). Among these, 724 proteins were exclusively identified by 1-D and only 4 proteins were identified exclusively on 2-D. Forty-four *T. rangeli* proteins revealed similarity to hypothetical proteins, but having EST and genomic support, were re-annotated as proteins of unknown function. Around 42% of *T. rangeli* proteins were associated to at least one Gene Ontology annotation, being transport and response to stress (BP), ion binding protein (MF) and cytoplasm (CC) the most abundant annotations per category. The combination of all techniques allowed the identification of stage-specific proteins during the *in vitro* differentiation process, which have been cloned, expressed and purified. The present proteomic data along with the parasite genome will constitute an important *T. rangeli* database, allowing the understanding of unknown aspects of this parasite biology.

RBP33 is an RNA binding protein which is regulated during the cell cycle and the differentiation of the bloodstream form *Trypanosoma brucei* (P223)*

Roman Trikin, Olivera Cirovic, Kapila Gunasekera, Nicholas Doiron, Christian Janzen, Torsten Ochsenreiter

Institute of Cell Biology, University of Bern, Switzerland Graduate School for Cellular and Biomedical Sciences, University of Bern, Switzerland Biocenter, University of Würzburg, Germany

Trypanosoma brucei is a single cell eukaryotic parasite causing human African sleeping sickness and Nagana in cattle. As part of the life cycle, the proliferative long slender parasite in the bloodstream of the mammalian host differentiates into the non-proliferative, cell cycle arrested short stumpy form, which is pre-adapted to life in the insect vector. The molecular mechanisms controlling the long slender to short stumpy differentiation are largely unknown. Using a combination of proteomics and transcriptomics we identified an RNA binding protein, RBP33, that we hypothesise to be involved in the control of cell differentiation. Overexpression of the protein in the BSF leads to cell cycle arrest in G2/M within 8-12 hours. During this time, overall mRNA and spliced leader RNA levels are significantly decreased while rRNA levels remain unchanged. Using biochemical and immunofluorescence microscopy assays we can show that RBP33 binds to polyA RNA *in vitro* and localises to the nucleus *in vivo*, respectively. We speculate that RBP33 is a polyA RNA binding protein important for regulation of total mRNA levels during the cell cycle and life cycle differentiation.

Otubain from *Leishmania chagasi* hydrolyzes K-48 linked ubiquitin and stimulates a pro-inflammatory response in murine peritoneal macrophages (P224)

Izabela M. D. Bastos1, Jhonata Pereira1,2, Clênia Azevedo1,2, Rafael Correa3, Flávia N. Motta1,2, Phlippe Grellier 4, Jaime M. Santana1, Kelly G. Magalhães3

1 Laboratory of Host-Pathogen Interactions, Department of Cell Biology, University of Brasília, Brasília, Brazil, 2 Faculty of Ceilândia, University of Brasília, Brasília, Brazil, 3 Laboratory of Immunology and Inflammation, Department of Cell Biology, University of Brasília, Brasília, Brazil, 4 Laboratoire d'Adaptation des protozoaires à leur environnement, MNHN, Paris, France.

Deubiquitinating enzymes (DUBs) play an important role in regulation of protein degradation, via 26S proteasome by deconjugation of ubiquitin (Ub) of labeled proteins, and in non-degradative processes. The otubain-1 is a DUB related to the regulation of lymphocytes anergy, an important immune modulator event. Deubiquitination in trypanosomatids demonstrates a promising alternative to search new therapeutic targets. This study aimed the enzymatic characterization of recombinant otubain from *Leishmania chagasi* (rOTULc), its localization within the parasite and its influence on the production of cytokines and lipid bodies in peritoneal macrophages. The recombinant WT and the F82S/F182S/L265P, F82S/L265P and L265P mutants were assayed on tetra ubiquitin K48- or K63-linked. The WT is shown to cleave Ub K48-linked and partially K63-linked. The L265P mutant showed only reduced activity on Ub K48-linked. The F82S/F182S/L265P and F82S/L265P mutants did not cleave either substrates. The macrophages (C57/Black6 mice) were stimulated with rOTULc for 24h and 72h. The cytokines profile was obtained by ELISA and the lipid bodies were counted. An increase of bodies and pro-inflammatory cytokines induced by rOTULc was observed in 24h and 72h, compared to controls, favouring a pro-inflammatory response. The immunofluorescence of rOTULc showed a vesicular pattern, closed to the kinetoplast. Our findings show that rOTULc cleaves preferentially tetra Ub K48-linked, a linkage associated to protein degradation by 26S proteasome suggesting an involvement in proteins preservation by avoiding their degradation. In addition, it is possible that rOTULc participates on immune response modulation of the host cells.

A directed evolution approach to reveal the immunoglobulin M binding site of Duffy-binding-like domains in malarial parasite-encoded proteins. (P225)*

Shona Moore1,3, Pat Blundell1, Dan Czajkowsky2 and Richard Pleass1
1 Liverpool School of Tropical Medicine, Pembroke Place, Liverpool, L3 5QA, UK; 2 Shanghai Institute of Applied Physics, Chinese Academy of Sciences; 3 Warwick Systems Biology Centre, University of Warwick, Coventry, CV4 7AL, UK

Duffy-binding-like (DBL) domains are essential to the function of proteins involved in critical interactions of the malarial parasite with its human host. These domains contained within numerous parasite antigens can interact with a wide array of host ligands leading to diverse disease outcomes. We recently characterised the interaction of DBL domains with human immunoglobulin M (IgM). At least ten DBL domains from *P. falciparum* isolates that cause diverse pathology including rosetting and placental malaria are known to bind human IgM, allowing the parasite to evade neutralization by antibody-mediated effector functions. In order to characterise the IgM-binding site of these DBL domains, we generated by PCR a library of ~150 novel DBL mutants using an error prone polymerase. Selected DBL mutants were expressed in CHOK1 cells and their affinity of binding to IgM investigated by ELISA and SPR-analysis. DBL mutants impacting on IgM-binding were then mapped onto known DBL crystal structures and an earlier model of the DBL-IgM interaction. Molecular dynamic simulations were used to further investigate amino acid interactions. These studies highlighted that amino acids outside of the proposed IgM binding site are critical for IgM-binding, suggesting that the structural model may be inaccurate, or that amino acids outside the proposed interaction site can also affect binding. DBL mutant data will be used to improve on the current model and identify further residues critical to binding. These will be investigated using site-directed mutagenesis. We hope to fully characterise the IgM-binding site of DBL domains using this updated structural model.

From Genome and transcriptome towards the development a new vaccine against leishmaniasis in Colombia (P226)

Omar Triana-Chávez, Daniel Urrea1, Juan P. Isaza3, Ana M. Mejía-Jaramillo1, Andrés Gómez1, José R. Ramírez-Pineda2, Juan Fernando Alzate3, and Omar Triana-Chávez1,
1 Biología y Control de enfermedades Infecciosas-BCEI, Universidad de Antioquia. 2 Grupo Inmunomodulación, Universidad de Antioquia. 3 Centro Nacional de Secuenciación Genómica, Medellín

The Leishmania parasite is an intracellular pathogen of the immune system targeting macrophages and dendritic cells. In America, about twelve million people are affected by leishmaniasis and 350 million people worldwide are at risk of infection. Leishmania panamensis is the most prevalent species in Colombia. To date there is no vaccine for leishmaniasis, and treatments have many complications. Therefore, It is necessary to make a rational search for vaccine candidates and new drugs, and this requires information of the genome and transcriptome of this parasite. As an approach to the identification of new vaccine candidate and the identification of species-specific genes, in the present

study we sequenced and assembled the genome of L. panamensis. Additionally, we sequenced and compared the amastigote and promastigote transcriptomes with the aim to identify amastigotes specific genes. We found that L. panamensis genome is 32 Mb in size, with a karyotype of 35 chromosomes as L. braziliensis. The G+C content is approximately 55.3%. Interestingly, the chromosome 30 was aneuploidy and the chromosome 31 was tetraploid. Comparative analysis with genomes of other species allowed us to select eight conserved genes highly immunogenic. We found 123 up-regulated and 127 down-regulated transcripts in amastigotes. The differential expression of some of these genes was verified by real-time PCR. We chose twenty of these genes to obtain recombinant protein and DNA vaccines to evaluate its antigenic potential. Further studies to evaluate the possible role of these genes in conferring protection in mice will be performed.

List of Attendees

Fathy Abdel-Ghaffar - Cairo University
Stephen Abolins - University of Bristol
Ibrahim Anka Abubakar - Usmanu Danfodiyo University, Nigeria
Fiona Achcar - University of Glasgow
Alvaro Acosta-Serrano - Liverpool School of Tropical Medicine
Emily Rebecca Adams - Liverpool School of Tropical Medicine
Salome Aeschlimann - University Bern, Switzerland
Andreia Albuquerque - Instituto de Higiene e Medicina Tropical (IHMT), Portugal
Jenna Alexander - Liverpool John Moores
Saeed Alharthi - Umm Al-Qura University, Saudi Arabia
Samuel Alizon - University of Montpellier, France
Abdulsalam Alkhaldi - Aljouf University
Hesham Al-Mekhlafi - University of Malaya
Mohammed Alruhaili - University of Salford
Waleed Al-Salem - Liverpool School of Tropical Medicine
Sam Alsford - London School of Hygiene & Tropical Medicine
Mohammed Alwabel - Qassim University, SAUDI ARABIA
Khalid Jamaan H Alzahrani - University of Glasgow
Seth Amanfo - University of Edinburgh
Jane Andre - Lancaster University
Stephen Attwood - State Key Laboratory of Biotherapy, China
Yvonne Azasi - University of Edinburgh
Noemi Bahamontes Rosa - GSK
Amna Arshad Bajwa - University of Veterinary & Animal Sciences Lahore, India
Rebecca Barber - University of Liverpool
Michael Barrett - University of Glasgow
Nicoletta Basilico - University of Milan
Geraldine Bastien - Edmund Mach Foundation
Jacob Baum - Imperial College London
Daniela Begolo - ZMBH, University of Heidelberg, Germany
Jerzy Behnke - University of Nottingham
Gus Bell - Trinity College Dublin
Hayley Bennett - BSP Meeting helper
Elodie Berdance - University of Bordeux, France
Martha Betson - Royal Veterinary College
Steve Beverley - Washington University, USA
Oliver Biliker - Session Organisers
Damer Blake - Session Organisers
Damer Blake - Royal Veterinary College
Tom Blundell - University of Cambridge
Brian Boag - Scottish Crop Research Institute
Cordula Boehm - BSP Meeting helper
Brenda Bondesen - Merial Limited, USA
Mark Booth - Wolfson Research Institute, University of Durham
Giovanna Boumis - Dept. Biochemical Sciences - "Sapienza" University of Rome
Reinhard Breitling - Jena Bioscience GmbH
Frederic Bringaud - Université Bordeaux Segalen, France
Andrew Briscoe - The Natural History Museum
Mathieu Brochet - Wellcome Trust Sanger Institute
Claire Butler - University of Oxford
Joanne Cable - Session Organisers
Susana Campino - Wellcome Trust Sanger Institute
Roberta Carloni - University of Edinburgh
Mark Carrington - Session Organisers
Eilidh Carrington - Liverpool School of Tropical Medicine
Lia Chappell - BSP Meeting helper
Antoine Claessens - Wellcome Trust Sanger Institute
Christine Elizabeth Clayton - ZMBH, University of Heidelberg, Germany
Dane Comerford - University of Cambridge
Sinclair Cooper - University of Edinburgh
Emmanuel Cornillot - Université Montpellier I, France
Ross Coron - University of St Andrews
Eloise Courtot - INRA
Janet Cox-Singh - St. Andrews University
George Cross - The Rockefeller University, USA
Jenny Crowe - University of Strathclyde
Zulma Cucunuba - Imperial College London
Krystyna Cwiklinski -
Viviane Da Silva - Universidade Federal de Minas Gerais, Brasil
Joel Dacks - University of Alberta, Canada
John Dalton - Queen's University Belfast
Filip De Bock - Zoetis, Belgium
Harry De Koning - University of Glasgow
Luis Miguel De Pablos Torro - University of Cambridge
Samuel Dean - University of Oxford
Godefroy Devevey - University of Edinburgh
Rebecca Devlin - University of Glasgow
Caroline Dewar - University of Edinburgh
Anaid Diaz - University of Cambridge
Rosario Diaz-Gonzalez - London School of Hygiene and Tropical Medicine
Kathrin Diederich - University of Heidelberg, Germany
Paul Divis - London School of Hygiene & Tropical Medicine
Tiago Donatelli Serafim - Welcome Trust Centre for Molecular Parasitology
Martin Donnelly - Liverpool School of Tropical Medicine
Richard Dorrell - University of Cambridge
Tim Downing - National University of Ireland, Galway
Lesley Drake - Imperial College London
Simon Draper - University of Oxford
Dorothea Droll - ZMBH, University of Heidelberg, Germany
Jean-Claude Dujardin - Institute of Tropical Medicine, Belgium
David Dunne - University of Cambridge
David Dunne - Session Organisers
Naomi Dyer - Liverpool School of Tropical Medicine
ThankGod E. Ebenezer - BSP Meeting helper
Martin M. Edreira - University Of Buenos Aires, Argentina
Jennifer Edwards - Aberystwyth University
Nicholas Ejeh - Liverpool School of Tropical Medicine
Egie Enabulele - Kinston University
Markus Engstler - Universität Würzburg, Germany
Astrid Erber - University of Oxford
Franco Falcone - University of Nottingham
Edward Farnell - BSP Meeting helper
Joe Farrimond - University of Edinburgh
Andy Fenton - University of Liverpool
Mark Field - Session Organisers
Matthew Fisher - Imperial College London
Thomas Fleming - Queen's University Belfast
Louise Ford - Liverpool School of Tropical Medicine
Suzanne Ford - University of Oxford
Bernardo Foth - Wellcome Trust Sanger Institute
Janaina Freitas - BSP Meeting helper
Matthias Fuegi - Swiss Tropical and Public Health Institute, Switzerland

Naomi Fulton - University of Edinburgh
Nicholas Furnham - London School of Hygiene & Tropical Medicine
Ondrej Gahura - Institute of Parasitology, Biology Centre ASCR
Jennie Garbutt - University of Edinburgh
Peter Geldhof - University of Ghent, Belgium
John Gilleard - University of Calgary, Canada
Eva Gluenz - Oxford University
Tim Goater - Vancouver Island University, Canada
Nick Golding - University of Oxford
Khurram Goraya - University of Health Sciences Lahore, India
Amy Goundry - University of Glasgow
Katy Graef - BIO Ventures for Global Health
Fabrice Graf - Swiss Tropical and Public Health Institute, Switzerland
Andrea Graham - Princeton University, USA
Bryan Grenfell - University of Princeton, USA
Raffaella Grimaldi - University of Dundee
Edmundo Grisard - Universidade Federal de Santa Catarina, Brasil
Maciej Grzybek - University of Life Sciences in Lublin, Poland
Keith Gull - University of Oxford
Tansy Hammarton - Session Organisers
Tansy Hammarton - University of Glasgow
Lance Hammerland - Merial Limited, USA
Matthew Hartfield - Centre National de la recherche scientifque (CNRS)
Marwa Hasby Saad - Tanta University, Egypt
amr hashem - Army forces, Egypt
Basripuzi Nurul Hayyan Hassan Basri - University of Glasgow
Emily Herman - University of Alberta, Canada
Roberto Hernandez - Instituto de Investigaciones Biomedicas UNAM, Mexico
Matthew Higgins - University of Oxford
Corinna Hiller - University of Heidelberg, Germany
Anders Hofer - Umea University, Sweden
Karl Hoffmann - University of Aberdeen
Cornelis Hokke - Leiden University Medical Center, Netherlands
Shazia Hosein - Royal Veterinary College
Tine Huyse - University of Leuven, Belgium
Eze Justin Ideozu - University of Salford
Joseph Igetei - University of Nottingham
Saher Islam - University of Veterinary & Animal Sciences Lahore, India
Andrew Jackson - University of Liverpool
Nusrat Jahan - GC University
Jo James - Cardiff University
Armando Jardim - McGill University
Kate Jones - University College London
Rebecca Jones - University of Liverpool
Eric Kalkman - University of Glasgow, Institute of Infection, Immunity and Inflammation
Oliver Kaltz - Université Montpellier 2, France
Gregory Karadjian - Muséum national d'Histoire naturelle, France
Paul Kaye - University of York
John Kelly - London School of Hygiene & Tropical Medicine
Louise Kerry - Imperial College London
Charlie King - CWRU, USA
Demelza Kingston - University of Glasgow
Ruth Kirk - University of Kingston
Agnieszka Kloch - University of Warsaw, Poland
Henrik Koch - University of Giessen, Germany
Linda Kohl - Muséum national d'Histoire naturelle, France
Julie Kovarova - University of Glasgow
Nina Krienitz - BSP Meeting helper
Sanjeev Krishina - St. George's University of London
James La Course - Liverpool School of Tropical Medicine
Bo Lai - BSP Meeting helper
Roz Laing - University of Glasgow
Poppy Lamberton - Imperial College London
Jean Langhorne - MRC National Institute for Medical Research
Jonathan Lappin - Queen's University Belfast
Mike Leahy - University of Oxford
Louis-Philippe Leroux - McGill University, Canada
John Lewis - Royal Holloway, Unversity of London
Michael Lewis - London School of Hygiene & Tropical Medicine
Sam Loker - University of New Mexico, USA
Ben Longdon - University of Cambridge
Christina Lorenz - BSP Meeting helper
Vincenzo Lorusso - University of Edinburgh
Julius Lukas - University of Southern Bohemia, Czech republic
Lukasz Lukomski - University of Liverpool
Amber Lynch - Lancaster University
Ashraf M. Ahmed - Session Organisers
Andrew Scott MacDonald - University of Manchester
Paula MacGregor - University of Edinburgh
Lorna Maclean - University of Dundee
Olivia Macleod - Cambridge University
Olivia Macloed - BSP Meeting helper
Luke Maishman - BSP Meeting helper
Rick Maizels - University of Edinburgh
Theresa Manful - University of Ghana, Legon
Paul Manna - BSP Meeting helper
Sujatha Manthri - University of Dundee
Neil Marks - University of Glasgow
Catarina de Almeida Marques - University of Glasgow
Jacqui Matthews - Session Organisers
Bella Maudlin - BSP Meeting helper
Aaron Maule - Queen's University Belfast
Johannes Mayer - University of Glasgow
Stella Mazeri - The Roslin Institute, University of Edinburgh
Hazel McDonald - University of Stirling
Glen McGugan - National Institutes of Health / NIAID
Paul McKean - Lancaster University
Maxine McKenzie - Kingston University
Carol McNair - University of Stirling
Tom McNeilly - Moredun Research Institute
Paul McVeigh - Queen's University Belfast
Isabela Mendes - University of Glasgow
Emma Louise Meredith - University of London
Catherine Merrick - Keele University
Louisa Alexandra Messenger - London School of Hygiene & Tropical Medicine
Paul Michels - University of Edinburgh
Adriana Erica Miele - "Sapienza" University of Rome
Michael Miles - London School of Hygiene & Tropical Medicine
Caroline Millins - University of Glasgow
Rachel Milne - University of Edinburgh
Katarzyna Modrzynska - BSP Meeting helper
Siti Nursheena Mohd Zain - University of Malaya

Karina Mondragon-Shem - Liverpool School of Tropical Medicine
Binny Mony - University of Edinburgh
Eric Mooring - University of Cambridge
Eric Morgan - University of Bristol
Brooke Morriswood - Max F. Perutz Laboratories
Monica Mugnier - The Rockefeller University, USA
Elizabeth Murchison - University of Cambridge
Nicholas Murphy - BSP Meeting helper
Janaina Nasciemento - BSP Meeting helper
Audun Helge Nerland - University of Bergen, Norway
Hatoon Abdullah M Niyazi - St. George's University of London
Victor Stephen Njom - The University of Sheffield
Billie Norman - Kingston University
Rachel Norman - University of Stirling
Fanny Nowacki - Aberystwyth University
Samson Obado - The Rockefeller University, USA
Torsten Ochsenreiter - University Bern, Switzerland
Mary Chiaka Oguike - London School of Hygiene & Tropical Medicine
Oluwashola Olaniyan - University of Edinburgh
Arnab Pain - KAUST, Saudi Arabia
Silvia Parapini - University of Milan
Emily Pascoe - Cardiff University
Laura Peachey - University of Liverpool
David Pigott - University of Oxford
Jason Pinger - The Rockefeller University, USA
Nicolas Pionnier - Muséum national d'Histoire naturelle, France
Lindsey Plenderleith - University of Edinburgh
Joaquin Prada - University of Glasgow
Christian Preusser - University of Giessen, Germany
Helen Price - Keele University
Stephen Price - University of Liverpool
Roger Prichard - McGill University
Cassandra Raby - University of Liverpool
Rajeev Rai - University of Greenwich
Srinivasa P S Rao - Novartis Institute for Tropical Diseases, Sinapore
Julian Rayner - Session Organisers
Andrew Read - Penn State, USA
Libby Redman - University of Calgary, Canada
Adam Reid - Wellcome Trust Sanger Institute
Jutta Reinhard-Rupp - Merck-Serona, Switzerland
Olivier Restif - University of Cambridge
Isabela Ribeiro - DNDi
Beth Richardson - BSP Meeting helper
Eva Rico Vidal - University of Edinburgh
Luke Roberts - Session Organisers
Luke Roberts - Imperial College London
Jean Rodgers - University of Glasgow
Isabel Roditi - University Bern, Switzerland
Ane Rodriguez Alejandre - GSK
Matthew Rogers - London School of Hygiene and Tropical Medicine
David Roos - University of Pennsylvania, USA
Clair Rose - Liverpool School of Tropical Medicine
Brice Rotureau - Institut Pasteur
Alex Rowe - University of Edinburgh
Gloria Rudenko - Imperial College London
Evelyn Rynkiewicz - University of Edinburgh
Ahmed M. Salman - The Jenner Institute - University of Oxford
Kathrin Schaten - University of Edinburgh
Bernd Schimanski - University Bern, Switzerland
Danae Schulz - The Rockefeller University, USA
Angela Schwede - BSP Meeting helper
Angela Schwede - University of Cambridge
Marco Sealey - Max F Perutz Laboratories
Karin Seifert - London School of Hygiene and Tropical Medicine
Darren Shaw - University of Edinburgh
Jennifer Shelton - University of Oxford
Lynn Sherrer - Cell Press
Tim Nicolai Siegel - University of Wurzburg, Germany
Eleanor Silvester - University of Edinburgh
Alexandra Sparks - University of Edinburgh
Anubhav Srivastava - University of Glasgow
Katharina Staerk - Royal Veterinary College
Henry Staines - St. George's University of London
Mario Steindel - Universidade Federal de Santa Catarina, Brasil
Jeremy Sternberg - University of Aderdeen
Russell Stothard - Liverpool School of Tropical Medicine
Karolina Subrtova - University of Southern Bohemia, Czech republic
Lalitha Sundaram - BSP Meeting helper
Jack Daniel Sunter - Oxford University
Colin Sutherland - London School of Hygiene & Tropical Medicine
Olivia Swann - University of Edinburgh
Balazs Szoor - University of Edinburgh
Melaku Tamene Asfaw - Addis Aabab University
Nick Taylor - Cefas
Martin Taylor - London School of Hygiene & Tropical Medicine
David Julian Timson - Queen's University Belfast
Paul Torgerson - University of Zurich, Switzerland
Moritz Treeck - MRC National Institute for Medical Research
Anna Trenaman - University of Dundee
Omar Triana-Chavez - Universidad de Antioquia, Colombia
Roman Trikin - University Bern, Switzerland
Mike Turner - Wellcome Trust
Elisabetta Ullu - Yale, USA
Vera Unwin - Liverpool School of Tropical Medicine
Michael Urbaniak - Lancaster University
Pedro Vale - University of Edinburgh
Angela Van Diepen - Leiden University Medical Center, Netherlands
Donelly van Schalkwyk - London School of Hygiene & Tropical Medicine
Sue Vaughan - Oxford Brookes University
Birgitte Vennervald - University of Copenhagan
Mark Viney - University of Bristol
Pegine Walrad - University of York
Arporn (Koi) Wangwiwatsin - BSP Meeting helper
Stephen Ward - Liverpool School of Tropical Medicine
Bonnie Webster - Imperial College London
David Wildridge - Institute of Parasitology
Mark Wilson - NIMR, London
Susan Mary Withenshaw - University of Liverpool
Dong Xia - University of Liverpool
Ya-Yi Yang - Leiden University Medical Center, Netherlands
Vyacheslav Yurchenko - University of Ostrava, Czech Republic
Magdalena Zarowiecki Zarowiecki - BSP Meeting helper
Andrea Zurita-Leal - University of Glasgow

Index

How do I find my poster spot?
Find your abstract title below at the end is a (P#) this is where your poster goes follow the signs to see the location. Each board has a P number on it. Items with a * next to the name are for the Student prize competition.

A

B

C

D

E

F

G

H

I

K

L

M

N

O

P

Q

R

S

T

U

V

W

Student Prize Voting Form

Prizes this year are
Best talk : Hp Laptop from the BSP, £200 in cash or books from CUP
2nd Placed talk £200 from Bio Med Central on behalf of Parasites and Vectors

Best Poster : Hp Laptop from the BSP, £200 in cash or books from CUP
2nd Placed talk £200 from Bio Med Central on behalf of Parasites and Vectors

Best student talks

1st choice:

Abstract Title/Session……..……

Presenter Name…….……

2nd choice:

Abstract Title/Session …….……..

Presenter Name…….……

Best poster

1st choice:

Abstract (P)No.……

Name…….

2nd choice:

Abstract (P)No.……

Name…….

Student Prize Voting Form

Prizes this year are
Best talk : Hp Laptop from the BSP, £200 in cash or books from CUP
2nd Placed talk £200 from Bio Med Central on behalf of Parasites and Vectors

Best Poster : Hp Laptop from the BSP, £200 in cash or books from CUP
2nd Placed talk £200 from Bio Med Central on behalf of Parasites and Vectors

Best student talks

1st choice:

Abstract Title/Session……..……

Presenter Name…….……

2nd choice:

Abstract Title/Session …….……..

Presenter Name…….……

Best poster

1st choice:

Abstract (P)No.……

Name…….

2nd choice:

Abstract (P)No.……

Name…….

Sunday 6th April						
12:00 -18:00	Registration					
19:00	**Reception Fitzwilliam Museum, Welcome address**					
Monday 7th April						
9:00-10.30	**Plenary lectures:** Tom Blundell, Bryan Grenfell and Keith Gull					
10:30-11:00	**Coffee break**					
	Session A West Road Recital	**Session B** West Road Lecture Room	**Session C** West Road Auditorium	**Session D** Robinson Auditorium	**Session E** Robinson Umney	**Session F** Robinson JCR
11:00 -12:30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
12:30-14:00	**Lunch and Naked Scientists**					
14:00-15.30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
15:30-16:00	**Coffee break**					
16:00-17:30	Tryp/leish	Apicomplexa	Tryp/Leish	Helminths	Ecology	Veterinary
17:30-18:00	**The Africa Experience:** David Dunne					
18:00-20:00	**Poster session and trade fair**					
20:00 on	**Young parasitologist party:** The Avery, Regent Street					

Tuesday 8th April						
09.00-9.30	**Plenary lecture:** David Roos					
9.30-10.30	**CA Wright medal lectures**-Alex Rowe and Michael Barrett					
10:3011:00	**Coffee break**					
	Session A West Road Auditorium	**Session B** West Road Lecture Room	**Session C** West Road Recital	**Session D** Robinson Auditorium	**Session E** Robinson Umney	**Session F** Robinson JCR
11.00-12.30	Tryp/leish	Apicomplexa	Evolution	Helminths	Communication	Veterinary
12:30- 14:00	**Lunch and social media**					
14.00-15.30	Tryp/leish	Apicomplexa	Evolution	Helminths	Ecology	Careers
15:30-16:00	**Coffee break**					
16.00 -17:30	Tryp/leish	Apicomplexa	Mid East	Helminths	Ecology	Veterinary
15:30-16:00	**Gala dinner: Robinson College**					
Wednesday 9th April						
9.30-10.30	**BSP Debate: Vector control - BSP AGM**					
10:30-11:00	**Coffee break**					
11.00-12.30	Tryp/leish	Apicomplexa	Mid East	Helminths	Ecology	Veterinary
12:30- 14:00	**Lunch and meeting close**					

The British Society for Parasitology is pleased to acknowledge the following who have either sponsored aspects of this meeting or who are exhibiting:

Parasites & Vectors

MALARIA JOURNAL

Royal Society Publishing

Caister Academic Press

CAMBRIDGE UNIVERSITY PRESS

ELSEVIER

Source BioScience LifeSciences

Newtonmicroscopes

dundeecell PRODUCTS

TwistDx

S T A R L A B

BRITISH SOCIETY FOR PARASITOLOGY
BSP