Società Chimica Italiana
Divisione di Spettrometria di Massa

FONDAZIONE EDMUND MACH

DIFFA23

DIRECT INJECTION FOOD FLAVOUR ANALYTICS

BOOK OF ABSTRACTS

Fondazione Edmund Mach

San Michele all'Adige (TN), Italy

20 - 22 September 2023

1st International Symposium on
Direct Injection Food Flavour Analytics (DIFFA)

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Research and Innovation Centre
Fondazione Edmund Mach
Via Mach, 1
38010 San Michele All'Adige (TN) Italy
phone +39 0461615427
fax +39 0461650872
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Proceedings of the DIFFA23 - 1st International Symposium on Direct Injection Food Flavour Analytics

Fondazione Edmund Mach – San Michele All'Adige (TN) Italy

20-22 September 2023

This book collects the conference proceedings of the 1st International Symposium on Direct Injection Food Flavour Analytics, held at the Fondazione Edmund Mach from 20th to 22nd September 2023.

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FOREWORD

Volatile organic compounds (VOCs), particularly flavour compounds, represent an invaluable noninvasive metric to follow the multi-faceted journey of food, from the farm to the fork and beyond, such as relating to the human microbiome after consumption or in addressing reduction strategies for food waste. VOCs thereby serve as a direct and swift means of measurement and notably act as a main driver of the perceived quality of food.

Mass spectrometry (MS) is an established yet increasingly pivotal tool in food and beverage characterization with a broad range of applications. When coupled with gas chromatography (GC), it stands as the predominant analytical method for exploring many aspects of food, from safety to traceability and nutritional aspects, and equally facilitates control measures in quality and process monitoring.

Recent remarkable advancements in both technology and methodology have paved the way for highly sensitive, specific, rapid, robust, and validated MS-based techniques that have become indispensable in food science and technology research and application. A subgroup of these technologies has been devised over the past two decades in the form of analytical approaches that enable the analysis of VOCs through direct injection. These methods have gained attention for their rapid, highly sensitive and high-throughput analytical capabilities.

A leading technology in this area is proton transfer reaction-mass spectrometry (PTR-MS), which has driven many innovative applications for direct flavour/food analysis. Commencing 2003, the University of Innsbruck, Austria, has organized a biennial event dedicated specifically to PTR-MS and its applications, including a focused session on food science and technology.

The **1st International Symposium on Direct Injection Food Flavour Analytics (DIFFA23)** was conceived with the backdrop of the PTR-MS conference but with a different aim, namely to embrace a broader community beyond PTR-MS uses, encompassing similar direct injection mass spectrometry (DIMS) technologies, such as atmospheric pressure chemical ionization-mass spectrometry (APCI-MS) and selected ion flow tube-mass spectrometry (SIFT-MS), with a primary emphasis on flavor compounds. It was also not exclusive to MS-based analytical techniques, but welcomed the inclusion of complementary non-MS approaches, such as solid-state sensors, fast gas chromatographic direct approaches and ion mobility spectrometry (IMS), amongst others, to ensure a wider reach and broader engagement. The meeting was established to foster scientific discussions of common interest and facilitate scientific collaborations. This book of abstract highlights the details of the event and contains the contribution summaries of both the oral and poster presentations.

The conference featured one plenary and four keynote lectures delivered by distinguished guests, as well as numerous invited and contributed talks and 25 poster presentations, with 97 attendees from different EU states, the USA, the UK, Israel and New Zealand. The event provided valuable insights into direct injection food/flavour analytics, with reviews from pioneering scientists who played key roles in developing and advancing DIMS methods in its early days, such as Andy Taylor, Patrik Španěl and Jean-Luc Le-Quéré, showcasing both historical developments and recent advancements in analytical performance and novel applications. Topics discussed included nose-space analysis of composite foods, rapid and high-throughput phenotyping, fermentation monitoring, both as an

innovative technological tool and for investigating the human microbiota, advanced data analysis and data mining tools. These are just a few examples of the themes explored during the conference.

Numerous partners contributed to the success of the event: the sponsors, whose engaging presentations and financial support sustained the quality of the meeting and ensured that the conference fees were kept to a minimum, as well as various supporting institutions and patronages. Special thanks go to the Fondazione Edmund Mach (FEM) for its scientific contributions and for hosting the conference at the Research and Innovation Centre, as well as the Division of Mass Spectrometry of the Italian Chemistry Society (DSM-SCI) for their organizational support and creation and hosting of the conference website. The invaluable support from these companies and institutions are further acknowledged through inclusion of their logos on the back cover of this book.

The conference started a fruitful exchange of results, ideas and issues amongst scientists working with direct tools to monitor VOCs in food science and technology, with broad attendance from sensory and applications scientists from academia and industry.

We would like to thank all those who, through their participation and support, made this event possible, which exceeded our most ambitious expectations.

Thank you all, and we look forward to seeing you at the next edition.

On behalf of the Scientific Committee

Franco Biasioli, Jonathan Beauchamp, Pat Silcock

CONFERENCE PROGRAM

20th September 2023

12.30-14.00 Registration and welcome buffet

Conference opening

14.00-14.10	Welcome addresses
	Fulvio Magni - *Società Chimica Italiana-Divisione Spettrometria di Massa*
	Mario Pezzotti - *Fondazione Edmund Mach*
14.10-14.20	Why DIFFA23?
	Franco Biasioli - *Fondazione Edmund Mach*
14.20-15.05	Plenary lecture: *DI-MS – A game changer for flavour research?*
	Andy Taylor - *University of Nottingham*

Session 1 | Unlocking Flavour with DIMS

Chairs: Pat Silcock & Nina Cleve

15.05-15.35	Jonathan Beauchamp - Fraunhofer Institute for Process Engineering and Packaging IVV
	The long and winding road: a flavoursome tale of PTR-MS
15.35-15.55	Graham Eyres - *University of Otago*
	What is Flavour and how can DIMS help untangle the puzzle?
15.55-16.15	Andreas Mauracher - *IONICON*
	Advantages of Next-Gen PTR-ToF instruments for food and flavour sciences

16.15-17.00 Tea break and poster session

Session 2 | DIMS in Health and Wellbeing

Chairs: Donatella Caruso & Eirini Pegiou

17.00-17.20	Josep Rupert - *Wageningen University & Research*
	Signalling volatile compounds in the human gut microbiota: new avenues offered by direct analytical methods.
17.20-17.40	Chris Mayhew - *University of Innsbruck*
	Real-Time Trace Analysis of Breath Volatiles using Proton Transfer Reaction Mass Spectrometry: implications for in-vivo flavour release measurements
17.40-18.00	Enrico Davoli - *Istituto Mario Negri*
	Direct analysis of sex-wellness products using a field deployable MS equipped with a Direct Sampling Atmospheric Pressure (DSAP) source
18.00-18.20	Corrado Di Natale - *University of Rome Tor Vergata*
	Direct injection mass spectrometry and gas sensors: a teacher-pupil relationship
18.20-18.40	Luca Cappellin - *University of Padua*
	Improved compound identification in direct VOC analysis using an EI&CI-TOFMS

19.00 Welcome cocktail - cloister of the monastery and historical cellar

21st September 2023

Session 3 | Linking DIMS Data to Sensory Perception

Chairs: Graham Eyres & Iuliia Khomenko

9.00-9.30	Jean-Luc Le-Quéré - *INRAE-CSGA Dijon* *Twenty years of Direct Injection Mass Spectrometry for aroma research in Dijon*
9.30-9.50	Catrienus De Jong - *Wageningen University & Research* *Exploring new in vivo and in vitro methods to integrate sensory and instrumental analysis to get insight and improve the flavour of plant-based food products during oral processing and drinking*
9.50-10.10	Markus Stieger - *Wageningen University & Research* *In vivo aroma release and sensory perception of composite foods*
10.10-10.20	Michele Pedrotti - *Wageningen University & Research* *Characterization of plant-based milks by combining sensory analysis with headspace and nose-space direct injection mass spectrometry*
10.20-10.30	Karina Gonzalez-Estanol - *Wageningen University & Research* *In vivo analysis of nose-space concentration by direct injection mass spectrometry to study the effect of chewing rate on aroma release during food consumption*
10.30-10.40	Laura Hill - *University of Nottingham* *Understanding the relationship between lipids, capsaicin and aroma release in confectionery*

10.40-11.10 Coffee break and poster session

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Chairs: Fulvio Magni & Caroline Perltier

11.10-11.30	Samo Smrke - *ZHAW School of Life Sciences and Facility Management* *Development of fast-GC PTR-MS method for coffee VOCs analysis*
11.30-11.45	Nina Cleve - *Fraunhofer Institute for Process Engineering and Packaging IVV* *Milk matters: Unraveling retronasal aroma release and perception of coffee by combining in vivo nosespace analytics with dynamic sensory methods*
11.45-12.05	Tomasz Majchrzak - *Gdansk University of Technology* *What happens when food goes into oil during deep frying? Monitoring the first minutes of frying using PTR-MS*
12.05-12.20	Gregory Schmauch - *Rational F&E GmbH* *Influence of product quantity, cooking parameter and flow tube pressure on the measurement with Sift-MS in a cooking oven*
12.20-12.40	Vaughan Langford - *Syft Technologies* *Application of SIFT-MS to chemical and sensory screening of packaging materials*

12.40-14.00 Conference group photo and lunch

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14.15-14.30	Matteo Tonezzer - *University of Cagliari* *PTR-MS as a tool to understand and improve the performance of electronic noses*
14.30-14.45	Andrea Warburton - *University of Otago* *Application of PTR-ToF-MS to monitor development of flavour in sourdough*
14.45-15.05	Paolo Redegalli - *Shimadzu Italia S.r.l.* *Characterization of isoflavones and its metabolites in foods by direct probe ionization mass spectrometer (DPiMS) with high resolution detection*
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Chairs: Riccardo Flamini & Michele Pedrotti

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16.45-17.05	Vittorio Capozzi - *Institute of Sciences of Food Production - National Research Council of Italy (CNR)* *DIMS techniques and the study on microbial VOCs in food: flavour attributes, fermentation monitoring and emerging trends*
17.05-17.20	Eirini Pegiou - *Wageningen University & Research* *Easy and fast detection of abnormal olive brine fermentation – A showcase of SPOTDETECT.*
17.20-17.40	Caroline Peltier - *INRAE* *Automatic pretreatment and multiblock analysis of flavor release and sensory temporal data simultaneously collected in vivo*
17.40-18.00	Ana Rita Monforte - *AFB INTERNATIONAL* *Modelling the kinetics of flavour formation & relaese as a function of ingredients addition in real food systems*
18.00-18.20	Pietro Franceschi - *Fondazione Edmund Mach* *Mining datasets from untargeted direct analytical methods: a data analyst point of view*
18.20-18.35	Mickael Le Bechec - *Institute of Analytical Sciences and Physico-Chemistry for Environment and Materials (IPREM)* *Volatile fingerprints of food thanks to the untargeted use of SIFT-MS raw data*

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22nd September 2023

Session 7 | Food Spoilage and Off-Flavour

Chairs: Catreinus de Jong & Brian Farneti

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10.00-10.15	Antonella Grosso - *University of Bolzano* *Monitoring autoxidation of vegetable oils by proton transfer reaction mass spectrometry*
10.15-10.30	Pedro Martinez Noguera - *University of Copenhagen* *Using PTR-ToF-MS to quantify microbial off-flavors geosmin and 2-methylisoborneol in water. Method development, performance assessment and comparison with established GC-MS methods*
10.30-10.45	Davide Papurello - *Turin Polytechnic* *Supporting sustainable energy production by PTR-MS: a review on the work accomplished on biofuel production from food waste to SOFC systems*
10.45-11.05	Rupert Holzinger - *Utrecht University* *Using SI traceable gas standards to improve the accuracy of untargeted PTR-MS measurements*

11.05-11.45 Coffee break and Poster Session

Session 8 | Floral, Biogenics and Phenotyping

Chairs: Rupert Holzinger & Vittorio Capozzi

11.45-12.05	Štefan Matejčík - *Comenius University* *Ion mobility spectrometry detection of plant hormones*
12.05-12.25	Brian Farneti - *Fondazione Edmund Mach* *DI-MS as high performing VOC phenotyping tool to support the horticultural production chain management*
12.25-12.40	Alberto Roncone - *Fondazione Edmund Mach* *Validation of gas chromatographic methods for the botanical characterization and authentication of lavender essential oil by stable isotope analysis of its organic volatile compounds*
12.40-12.55	Eugenio Aprea - *University of Trento* *Contribution of volatile organic compounds to multifloral honey flavor*
12.55-13.15	Daniele Zatta - *University of Padua* *Comparative analysis of VOC purification techniques in complex cooking emission: adsorption, photocatalysis and combined systems.*

13.15-13.30	Closing remarks Fulvio Magni - *Società Chimica Italiana-Divisione Spettrometria di Massa* Franco Biasioli - *Fondazione Edmund Mach*

13.30 Farewell buffet

LIST OF CONTRIBUTIONS

P.01 Effect of different carbon sources on fermentation volatile organic compounds (VOCs) profile by *Levilactobacillus brevis* WLP672 using proton transfer reaction-time of flight-mass spectrometry (PTR-ToF-MS)

Sarathadevi Rajendran[1,2,3]*, Iuliia Khomenko[2], Patrick Silcock[1], Emanuela Betta[2], Franco Biasioli[2], and Phil Bremer[1]

[1]Department of Food Science, University of Otago, P.O. Box 56, Dunedin 9054, New Zealand
[2]Sensory Quality Unit, Research and Innovation Centre, Fondazione Edmund Mach, Trento, Italy
[3]Department of Agricultural chemistry, Faculty of Agriculture, University of Jaffna, Sri Lanka

*pat.silcock@otago.ac.nz

Summary: The volatile organic compounds (VOCs) produced by the lactic acid bacterium, *Levilactobacillus brevis* WLP672, growing on a range of carbon sources in a defined nutrient medium under a range of fermentation conditions (time, and temperature) were assessed using proton transfer reaction-time of flight-mass spectrometry (PTR-TOF-MS). This study will help to identify plant-based substrates that could be used to produce meat, or dairy flavours via bacterial fermentation.
Keywords: defined nutrient medium, plant-based substrates, volatile organic compounds (VOCs), lactic acid bacterium, meat, or dairy flavours

1 Introduction

Plant-based diets have gained popularity as consumers strive to reduce the environmental impact of their diet while also improving their health and addressing animal welfare concerns [1]. Despite the abundance of meat and dairy analogues, these analogues still lack the overall sensory appeal of their traditional counterparts [2] owing to the challenges associated with obtaining a realistic meat or dairy flavour [3]. Bacterial fermentation is considered to be a cost-effective means of generating desired flavour compounds from plant-based substrates [4]. However, the wide range of substrates present in plants makes it challenging to understanding how individual components impact on volatile organic compound (VOC) production. To simplify this, a defined medium can be used to better understand how compounds that may be present in plants impact on VOC production. The current study used a defined nutrient medium to determine the effect of different carbon sources (either glucose, fructose or citrate) on the VOCs produced during fermentation by *Levilactobacillus brevis* WLP672 (*Lev. brevis* WLP672).

2 Experimental design

The defined medium (5mL) containing peptone, sodium acetate, mineral salts, vitamins, and a carbon source of either glucose (AG), fructose (AF), or citrate (AC) was added to sterile headspace vials (20 mL). Vials were inoculated with 0.05 mL of a *Lev. brevis* WLP672 culture (1 ×1011 CFU/mL) and N_2 gas was flushed at a rate of 10 mL/min for 20 min into each headspace vial to obtain an anaerobic environment. Vials were incubated in sample trays for 14 days at either 25 or 35 °C in an autosampler specially adapted for proton transfer reaction-time of flight-mass spectrometry (PTR-ToF-MS). The VOCs produced during fermentation were analyzed at 0, 7, and 14 days by PTR-ToF-MS. To aid VOC identification, samples were also analysed at day 14 using solid phase microextraction-gas chromatography- mass spectrometry (SPME-GC-MS).

3 Results

A total of 267 mass peaks from the raw PTR-ToF-MS spectra were reduced to 105, after removal of isotopologues and mass peaks that were not significantly ($p>0.05$) different from the baseline. Tentative identification (t.i.) of each mass peak was based on its exact mass, supported by SPME-GC-MS identification and literature data. Among the 105 mass peaks, 83, 72 and 68 mass peaks were significantly ($p<0.05$) differentiated based upon carbon source, temperature, and defined media*temperature interactions, respectively. The identified VOCs were classified as being either acids, alcohols, aldehydes, esters, furans, ketones, or sulphur compounds. The concentrations of two representative dairy flavours after 7 days of fermentation, m/z 45.033 (t.i. acetaldehyde) (Figure 1a) and m/z 89.060 (t.i. butyric acid/ethyl acetate) (Figure 1b) were significantly ($p<0.05$) higher at 35 °C in the AG defined medium than at 25 °C. These two compounds were also higher in the AG defined medium than in either the AF or the AC defined medium at both 25 and 35 °C. After 7 days of fermentation, the concentration of a representative meat or dairy flavour, m/z 49.011 (t.i. methanethiol) was significantly ($p<0.05$) higher in the AG defined medium at 35 °C than in all other treatments (Figure 1c). In addition, m/z 49.011 in the AF defined medium at 35°C was significantly higher ($p<0.05$) than in the AG and AF defined media at 25 °C, and in the AC defined medium at 25 and 35 °C.

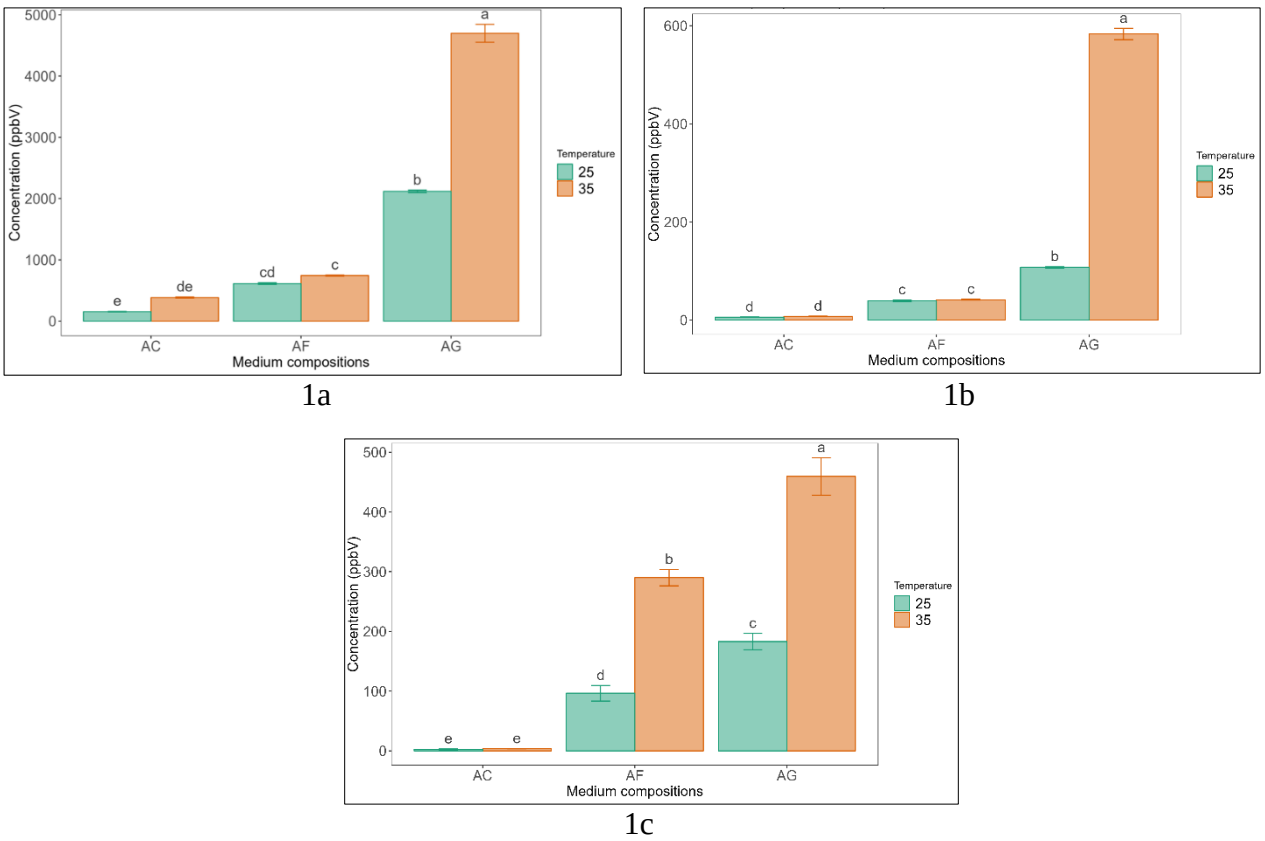

Figure 1. *Mean concentration (ppbV) of m/z 45.033 (t.i. acetaldehyde) (1a), m/z 89.060 (t.i. butyric acid/ethyl acetate) (1b), and m/z 49.011 (t.i. methanethiol) (1c) across the different defined nutrient medium compositions (AG, AF, and AC) at 25 and 35 °C after 7 days of fermentation by Lev. brevis WLP672. Different letters represent significant differences between the different medium compositions according to Tukey's test at $p<0.05$.*

5 Conclusion

VOCs produced by *Lev. brevis* WLP672 via fermentation in a defined nutrient medium were strongly influenced by carbon sources and the fermentation conditions (time and temperature). Overall, the defined medium containing glucose generated higher concentrations of VOCs of interest during fermentation at 35 °C compared to media containing either fructose or citrate. The results from this study will help to determine how to target the production of fermentation VOCs that mimic meat or dairy-like flavours by supplementing the substrate composition or adjusting the fermentation conditions. The knowledge gained through this research could be used to enhance the production of desirable VOC on an industrial through the fermentation of plant-based substrates.

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