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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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

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	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

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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

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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*
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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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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

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Chairs: Catreinus de Jong & Brian Farneti

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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

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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

O.16 Contribution of volatile organic compounds to multifloral honey flavor

Eugenio Aprea[1]*, Danny Cliceri[1], Emanuela Betta[2], Flavia Gasperi[1]

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[2]Research and Innovation Centre, Fondazione Edmund Mach, San Michele All'Adige, Italy

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Summary: With the scope of exploring flavor determinants of multifloral honeys, the VOC profile of multifloral honeys from Trentino area were acquired by SPME/GC-MS and associated with the sensory descriptive profiles obtained by Check-all-that-apply (CATA) method generated by a trained panel of honey experts.
Keywords: SPME/GC-MS, CATA, Multifloral honey

1 Introduction

Multifloral honey is derived from the nectar of multiple natural sources, collected by honeybees from various flowers and plants, resulting in a blend of different floral flavors. From a chemical-physical standpoint, multifloral honey includes honeys that don't fit within limits set for "monofloral" honeys, forming a continuum that makes it hard to distinguish characteristic groups. The high variability of botanical species in multifloral honey causes its "downgrading," but also grants it complexity and unique sensory quality. The variety and quantity of nectar sources in multifloral honey are closely linked to the territory and seasons. For instance, multifloral honey from Trentino blends temperate, sub-Mediterranean, Mediterranean, and Alpine climates, creating a diverse botanical variety. Understanding whether this combination generates distinctive characteristics for a specific area is relevant for local product promotion.
From the flavor point of view, multifloral honey can be considered the maximum expression of the flora of a territory, often resulting in unique and often unrepeatable combination of sensory characteristics. These peculiarities can be objectified with sensory science approaches that make it possible to obtain the descriptive profile of a food. In a broader sense, the enhancement of multifloral honey can find a more complete vision by studying the volatile organic compounds (VOC) responsible for the differ flavor components. The aim of the study was to identify the VOCs mainly involved in the odor and flavor perception of multifloral honey. In this first attempt we focused on honey flavor macro categories.

2 Experimental

Samples: Thirty-six multifloral honeys produced in 2021 collected from the Trentino (Italy) representative of the different mountain areas with different botanical characteristics.
VOCs profile: VOCs were measured by SPME/GC-MS. 1 g of honey was mixed with 1 mL of distilled water and 0.5 g of NaCl within a 20 mL GC vial and spiked with 2-octanol. SPME and GC-MS details can be found in [1]. Data were reported as the relative amount of 2-octanol used ad IS.
Sensory profile: After proper training, 47 judges described the odors by smelling and the flavors by tasting the samples (14 g of honey in a 40 ml glass jar) using CATA method [2] with sensory attributes from the honey sensory wheel [3] and evaluated their category representativeness. The evaluations were carried out remotely and samples were provided anonymously and in randomized order.

Data analysis: Attributes from CATA were submitted to Cochran Q test to identify significant discriminating descriptors. Spearman correlation coefficient was applied to study associations between category representativeness and attributes. Multivariate visualization was obtained through Multi Factor Analysis applied on discriminating descriptors. Data from SPME/GC-MS were pre-processed by a Log transformation, mean-centered and scaled to pareto scaling. CV-ANOVA was performed to assess the reliability of PLS predictive models cross-validated [4]. The coefficients were used to assess the contribution of the single volatile compounds to the model of each sensory descriptor. Multivariate analyses were performed by SIMCA 17.2 software and R software v 3.1.1.

3 Results

Sensory data: 49 sensory attributes out of 55 were found to be significantly discriminative. The descriptors with a positive contribution on category representativeness (Rho > 0.7) were related to the odor and flavor of the Fruity family, the Vegetable, the Animal, the Acid taste, the Astringent sensation, the light Amber color, and a Fluid consistency.

VOCs data: From Honey chromatograms 118 peaks have been extracted of which 108 have been identified while 10, present clearly in some of the honeys, are reported as unknown. Fourty-four compounds were present in all the analyzed honeys and 18 compounds were present in at least half of the samples. The chemical class mainly represented is that of the terpenes with at least 47 compounds (at least 2 of the unknown present typical fragments of terpenes). The other compounds, reported in decreasing order, belong to the following chemical classes: alcohols (16), aldehydes (10), esters (8), acids (7), ketones (6), furans (4), norisoprenoids (3), N-compounds (3), hydrocarbons (2) and a S-compound and the cyclohexyl isothiocyanate.

Association models. To identify the association between VOCs and the sensory descriptors (odor and flavor attributes), OPLS regression models were built and tested by CV-ANOVA ($\alpha<0.001$) and the coefficients of these models were used to identify the contribution of the VOCs to the different sensory descriptors in honeys (Table1).

Table 1. *Main associations between sensory macro categories and VOCs. o- odor, f- flavor*

Attribute	**Main associated VOCs**			
o-Floral	4,5-Dimethylfurfural	Heptanal	Nonanal	Lilac Aldehydes (A,B,C,D)
o-Warm	Ethyl hexadecanoate	Ethyl tetradecanoate	3-Methyl-3-buten-1-ol	Lavender lactone
o-Aromatic	Ethyl tetradecanoate	Ethyl hexadecanoate	Isoamyl acetamide	Thymol
o-Chemical	Ethyl tetradecanoate	Ethyl hexadecanoate	8-p-Menthen-1,2-diol	Isoamyl acetamide
f-Floral	4,5-Dimethylfurfural	p-Cymen-8-ol	Lilac Aldehydes (A,B,C,D)	(Z)-Linalool oxide (furan)
f-Fruit	Terpenediol I	UnknownH	UnknwonF	Ipomeanol
f-Warm	Ethyl hexadecanoate	Ethyl tetradecanoate	3-Methyl-3-buten-1-ol	Borneol
f-Aromatic	Isoamyl acetamide	Benzyl Alcohol	Ethyl tetradecanoate	8-p-Menthen-1,2-diol
f-Chemical	Ethyl tetradecanoate	8-p-Menthen-1,2-diol	Thymol	Ethyl hexadecanoate

For example, among the main contributors to the o-Floral, we found heptanal, described as fresh, green, citrus-like [5], nonanal, described as waxy, aldehydic, citrus, green lemon peel like and

cucumber fattiness [5] and the 4 stereoisomers of lilac aldehyde that are characterized by flowery odors [6]. The sensory categories of o-Warm, o-Aromatic and o-Chemical partial overlap and encompass a broad range of more specific attributes. Their common characteristics are supported by the presence of the two esters ethyl tetradecanoate and ethyl hexadecanoate which contribute sweet, waxy, fruity, creamy, and balsamic notes [5]. However, the differentiation among the three categories is defined by the combinations of specific compounds, mainly belonging to the terpene class.

4 Conclusions

In this study, we assessed the qualitative impact of volatile organic compounds on odor and flavor macro attributes in multifloral honey. The identified associations align with the sensory influence of individual compounds, whose combination give rise to the rich flavor profile of the honeys. We believe that this approach can be expanded to sensory subcategories to encompass more precise sensory attributes, thereby enhancing the correlation between odor/flavor descriptors and chemical compounds.

References

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