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Welcome, by the Chairs of ISEAC-41

Welcome to the 41st International Conference on Environmental & Food Monitoring, ISEAC-41, in Amsterdam - The Netherlands, to continue with us the conference series under the umbrella of the International Association of Environmental Analytical Chemistry (IAEAC).

The conference focuses on innovations in analytical techniques covering the environment-food continuum. After a period of pandemic-related absence, ISEAC-41 provides a great platform to highlight the latest scientific and technological achievements, exchange of out-of-the-box ideas, meet with vendors and discover the latest analytical developments, strengthen existing collaborations and, last but not least, expand your scientific network and initiate new scientific adventures.

Sustainability and planetary boundaries have made their appearance in the public debate, and integrated solutions for a better living environment and a healthy life are needed now more than ever. At ISEAC-41, challenges in both environmental and food monitoring and analysis will be addressed, and trends and future perspectives will be demonstrated.

The daily conference program includes renowned keynote speakers that will provide exciting insights into their groundbreaking work, while at the same time, special attention will be given to young scientists, providing them with a platform to share their findings and discuss ways forward. Poster sessions provide an excellent opportunity to get inspired, foster collaborations and discuss burning questions. A continuous exhibition gives the opportunity to connect to a great selection of companies, big and small, providing the latest laboratory equipment and solutions.

And obviously, there will be plenty of opportunities to enjoy the city of Amsterdam!
We warmly welcome you in Amsterdam, for an inspiring and exciting conference in a friendly and open atmosphere in one of the most beautiful European capitals!

Marja Lamoree
Hans Mol

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KeyNote Presentations

The present and future use of monitoring data in EFSA's human health risk assessment of chemicals

B. Dujardin, C. Cascio, S. Levorato, A. Papanikolaou

European Food Safety Authority

The European Food Safety Authority (EFSA) provides scientific advice to the European Commission, the European Parliament and the EU Member States on risks associated with the food chain. Since its establishment in 2002, EFSA has acquired robust expertise in the field of dietary exposure, which is one of the key components of chemical risk assessment in food. This has resulted in the building of the Comprehensive European Food Consumption Database and a wealth of chemical monitoring data in food have been collected for a wide range of chemical compounds, including pesticide residues, chemical contaminants, veterinary medicinal products, food additives and nutrients. In addition, to facilitate the execution of dietary exposure calculations as part of EFSA's regulatory processes, a range of open access tools was made available to both internal and external users.

Whereas the tools, methods and processes developed by EFSA so far were primarily designed for calculating dietary exposure to single chemicals, consumers are exposed to a multitude of chemicals from a variety of sources. To allow for the integration of chemical mixtures and multiple sources of exposure into daily risk assessment, EFSA needs to explore new data streams and stay on the look-out for new innovative concepts. A first roadmap for action on Risk Assessment of Combined Exposure to Multiple Chemicals has already been issued and a second roadmap for action on Advancing Aggregate Exposure to Chemicals in the EU is currently under development. Furthermore, EFSA has initiated a project investigating new opportunities regarding monitoring and surveillance data for chemicals.

This oral presentation will provide an overview of the ongoing initiatives of EFSA in this domain, and provide insight on how EFSA intends to probe into new surveillance data sources ranging from survey data, such as market shares, to analytical data, such as human biomonitoring.

From target to non-target screening of chemical mixtures in water - in situ, frequently, hazard-based, retrospectively

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Synthetic chemicals are everywhere in our daily life and many end up in the aquatic environment. Chemicals with hazardous properties can pose a threat to the environment, as well as exacerbating planetary crises such as ecosystem degradation and biodiversity loss. The situation is challenging for water quality assessment, where usually only a selected number of compounds are monitored (e.g. within the Water Framework Directive) to evaluate associated risks. Recently, the European Commission introduced the Green Deal as a vision toward a sustainable society and markets. The key elements for reducing pollution are the "Zero Pollution Ambition for a toxic-free environment", the "Chemicals Strategy for Sustainability" and the action plan "Towards Zero Pollution for Air, Water and Soil". A crucial element for the success of these new policies is the availability of comprehensive monitoring data.

Improvements in chemical analytical methods, such as high resolution mass spectrometry (HRMS), combined with data science and hazard prediction, has opened new opportunities for addressing combined exposure to chemical mixtures from diverse sources over time. The International River Commissions for the Protection of the Rhine and the Danube are currently evaluating early warning systems employing HRMS. Several Member States have initiated the development of HRMS databases for real-time and retrospective data analysis. Non-target screening using HRMS is also a key component of environmental monitoring within the new Partnership for the Risk Assessment of Chemicals (PARC). In addition to presenting the overarching framework, this presentation will showcase examples of in situ, more frequent, hazard-based and retrospective monitoring of a broader spectrum of compounds in aquatic systems. Comprehensive target, suspect and non-target screening of chemicals with HPLC coupled to tandem HRMS, alongside diverse prioritization approaches will be illustrated. Furthermore, challenges regarding harmonization of these methods to facilitate regulatory uptake will be discussed (Hollender et al., 2023).

References:

Hollender et al. (2023) NORMAN guidance on suspect and non-target screening in environmental monitoring, Env Sci Europe https://doi.org/10.1186/s12302-023-00779-4

Innovative sampling and suspect / non-targeted screening coupled to effect direct analysis approaches as a support to next generation of risk assessment: the Task 4.3 component of the Partnership for the Assessment of Risk from Chemicals (PARC)

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Human populations are exposed on a daily basis to multiple chemicals via various routes, including the environment, food or lifestyle, among which major concerns are related for instance to persistent organic pollutants (POPs), endocrine disrupting chemicals (EDCs) and/or chemicals of emerging concern (CECs). At a scientific level, the main challenge associated to the detection of these extended real life chemical mixtures is to develop new methodological strategies to document the reality of such exposure and its related health impact. At a regulatory and policy level, the main challenge is to develop early warning capability to handle these chemicals of both exposure and toxicological concern through biomonitoring program and further risk assessment process. Suspect and non-targeted screening (SS/NTS) approaches coupled to Effect-Directed Analysis (EDA) represents a major advance in the field in that respect, and appears as a relevant strategy for a better integrated exposure and hazard characterization. New sampling methods at individual level (e.g. silicone wristbands and/or dried blood spots) or at community level (wastewater based epidemiology) are also a matter of growing interest. This field is however still facing significant challenges that entail major analytical developments related to each step of the workflow. On one hand, harmonisation measures are required to achieve better consolidation and comparability of data generated from various studies, especially regarding their further use in a regulatory and support to policy context. On the other hand, this evolving field requires preserving flexibility in order to maintain its capacity in discovery and exploratory research. In the continuity of the previous HBM4EU initiative (H2020, 2017-2022), the Task 4.3 component of the PARC consortium (Horizon EU, 2022-2029) was tailored to address a number of these challenges, through a structuration based on both transversal activities aiming to foster permeability and alignment between the environmental, food, and HBM communities with regard to some common issues including QAQC or data processing workflow, and several complementary proof-of-concept aiming to assess and better define the applicability of these new approaches in various contexts related to each of these 3 silots. The presentation will describe those different activities developed within this Task co-led by INRAE and VU and associating 67 institutes from 17 countries.

Chemical cocktails threaten the environment and human health: it's all in the mix!

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An overwhelming number and diversity of organic chemicals are in daily use and enter the environment during or after use and disposal. Chemical pollution is an increasing threat to our environment, to wildlife and to people. The impact of chemical pollution will be amplified by population growth and climate change. However, conventional chemical monitoring programs have been criticized on the basis that they cannot include the full range of chemical pollutants that could occur in the environment including transformation products, and they do not account for the combined effects of mixtures of chemicals. Bioanalytical tools may therefore complement chemical analysis for cost-efficient and comprehensive (bio)monitoring. Bioanalytical tools are *in vitro* cell-based bioassays that target specific mechanisms of toxicity and give a measure of the toxicity of mixtures of known and unknown chemicals. Bioanalytical tools provide measures of the cumulative effects of mixtures of chemicals that exhibit the same mode of toxic action plus they can give a measure of the cytotoxicity of all chemicals.

One of the biggest challenges for the application of in vitro bioassays is the comprehensive extraction of organic chemicals from complex organic matrices that are lipid-rich such as adipose tissue, or whale blubber or rich in proteins such as fish, milk or blood but even the natural organic matter in surface water might pose a challenge. A smart combination of passive equilibrium sampling for hydrophobic pollutants together with solid-phase extraction for more hydrophilic and ionizable organic chemicals leads to a defined extraction of blood and similar matrices. Using case studies on the continuum from wastewater/surface water via drinking water and food (fish and milk) to biomonitoring of human tissues and blood I will illustrate how a combination of chemical analysis and bioanalytical tools in conjunction with mixture modelling can help to understand which fractions of the chemical pollution are known and which are unknown. Improved detection of the presence of mixtures of chemicals in people informs chemical risk assessment and management options.

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Advances in microplastic exposure, fate, effects and risks

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The assessment of microplastics in the environment represents a unique challenge to researchers. Microplastics in the environment are a complex mixture of polymer types, additives, shapes, sizes, and other defining characteristics (Koelmans *et al*., 2022). In order to describe potential risks associated with microplastics, researchers need to be able to distinguish between effects caused by microplastics from natural and other anthropogenic particles and chemicals. Microplastics also have the added challenge of potential effects being caused by the physical interaction with the microplastic particle (e.g. shape and size) or by interactions with the microplastic's chemical composition. As for shape and size, this is relevant to the 'food dilution' effect mechanism of MP (Koelmans *et al*. 2020). This makes it necessary to develop analytical techniques that can quickly and routinely estimate the volume of particles (Barchiesi *et al.* 2023). By assessing potential mode(s) of action for toxicity and their dose responses, probability density functions for particle characteristics, e.g. particle volume, and advanced statistical analyses can be used to determine the potential risks of any mixture of microplastics (Koelmans *et al*., 2023). This framework is applicable to both ecological and human health risk assessments. These unique set of challenges will be discussed, and the latest risk assessment frameworks will be presented to the attendees.

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Advancing Green and Sustainable Analytical Chemistry for Environmental and Food Analysis

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Green and sustainable analytical chemistry encompasses more than just evaluating the environmental impact of analytical methods; it emphasizes their broader societal implications [1]. This necessitates the development of innovative analytical techniques that contribute to the attainment of the 17 Sustainable Development Goals, including critical concerns such as food safety and security, biodiversity preservation, and efficient utilization of natural resources.

As an analytical chemist, you have a vital role in (i) developing higher-quality analytical methods, (ii) pursuing greener alternatives, and (iii) prioritizing socially relevant projects. Our research has demonstrated the efficacy of utilizing supercritical carbon dioxide (scCO2) and green co-solvents like ethanol, ethyl lactate, and water for rapid extraction of target compounds from complex plants, food, and environmental samples [2]. Likewise, we have achieved rapid separations with exceptional efficiency by employing supercritical fluid chromatography (SFC) with $scCO_2$ mixed with methanol as a co-solvent [3]. In this lecture, we will showcase the extractability, selectivity, and comprehensiveness of supercritical fluid extraction (SFE) using mixtures of carbon dioxide, ethanol, and water in the analysis of seaweed [4]. Seaweed presents an intriguing marine food source with significant potential as it requires no arable land, irrigation, fertilizers, or pesticides. Our findings demonstrate that SFE enables high extractability and selectivity when targeting health-beneficial antioxidant compounds in seaweed while effectively minimizing the co-extraction of toxic metals, such as arsenic. Furthermore, we will provide examples from our food and environmental analysis projects to illustrate the use of SFC with charged aerosol detection and high-resolution mass spectrometry to enable both quantification and identification of unknown compounds, even in the absence of chemical standards [5-6].

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Strategies for characterizing the consumer's chemical exposome

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Chemical contaminants, whether of natural or synthetic origin, produced intentionally or not, are all hazards likely to enter the food chain. Depending on their toxicity and levels of exposure, they can represent a risk to consumers, and assessing and controlling these risks is more than ever a public health priority. As awareness of the multitude of substances has grown, so has research into the detection and characterization of these contaminants. Laboratories are deploying two strategies to address this particular issue: (i) one focuses on substances that are already known or have recently been described, with the aim of developing high-performance analytical approaches to objectively identify the presence of these contaminants in foodstuffs and measure their concentration levels, in order to help characterize consumer exposure; (ii) the other explores emerging hazards in a more global way, using research strategies based on particular chemical motifs (e.g. halogens), specific effects (e.g. involving metabolomics) or the modelling of probable structures. The innovative analytical strategies associated with these two approaches will be detailed to illustrate the identification of emerging hazards in the food chain with the aim of extending knowledge of the chemical exposome of consumers.

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Endocrine disruptors in freshwater: policy perspectives by the OECD

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Endocrine disrupting chemicals are contaminants of emerging environmental and healthconcern that have been detected in freshwater, wastewater and drinking water. They interfere with the endocrine system in humans and wildlife. Their presence in water raises concerns for the integrity of ecosystems and biodiversity. Member countries of the Organisation for Economic Co-operation and Development (OECD) have therefore intruduced several policy actions and monitoring programmes to address the issue of endocrine disruptors.

This presentation will share the highlights of the OECD (2023) report *Endocrine Disrupting Chemicals in Freshwater: Monitoring and Regulating Water Quality*. This report calls for a better understanding, better monitoring, and policy actions to prevent and remedy emerging concerns around endocrine disruptors in the environment. The report builds on scientific knowledge and a range of case studies from OECD countries to provide practical examples and concrete policy actions.

A monitoring toolbox combining different methods, for instance traditional chemical analysis, bioanalytical methods and non-targeted screening, is well-equipped to inform regulatory decisions. While academia has been a frontrunner in applying novel water quality monitoring methods, notably bioassays but also high-resolution mass spectrometry (HRMS), policymakers are facing several barriers in the roll-out of such methods for regulatory purposes. Some of the barriers are i) access to method suppliers, ii) costs, iii) laboratory infrastructure, and iv) setting threshold values for new methods.

The vast range of sources and entryways of this group of chemicals into the environment makes designing policy interventions even more complex. Policy instruments that address the life cycle of endocrine disruptors – from source, to use, to end-of-pipe - can reduce their presence in water. The report also proposes tools and regulations that respond to the negative effects of endocrine disruption, even if the culprit chemical is still unknown.

References:

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Advances on the identification, monitoring, and prioritization of persistent, mobile and toxic (PMT) substances threatening water resources

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Water resources are becoming increasingly contaminated with substances considered persistent, mobile and toxic (PMT) or very persistent and very mobile (vPvM). PMT/vPvM substances are those which do not breakdown easily in the environment (persistent), are transported relatively rapidly through soil and groundwater (mobile) and are in some cases toxic. Their irreversible accumulation in the environment collectively allows them to fulfil the criteria of a planetary boundary threat. The most well known PMT/vPvM substances are short-chain perfluoroalkyl substances (PFAS), such as trifluoracetic acid, as well as melamine and 1,4-dioxane. But this is just the tip of the iceberg: there are many more, including a large portion of transformation products that remain unidentified. Advanced water treatment methods like ozonation or activated carbon filtration are often ineffective, forcing some water producers to invest in nanofiltration and reverse osmosis. As part of a larger strategy to address the threat posed by these the substances, the EU's CLP Regulation introduced new hazard classes PMT and vPvM in April 2023.[1] The presentation will present the history of the concern and characterization of PMT/vPvM substance contamination, which ultimately led up to the introduction of new hazard classes. This will be followed by different ways of identifying and prioritizing PMT/vPvM substances[2], including i) assessing the PMT/vPvM hazard; ii) assessing the emission likelihood; iii) identifying the analytical and monitoring gaps; iv) the remediation gaps and v) the exposure level. The talk will conclude with further areas of research for better identifying and monitoring diverse PMT/vPvM substances and PFAS in the environment, and strategies towards zero pollution from these substances[3].

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New Challenges and Opportunities in Food Analysis

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Food analysis is an evolving field that has been influenced by various factors, including advancements in analytical techniques and technology, changes in regulatory requirements, and developments in food technology. Recently, there has been a significant focus on the introduction of alternative protein sources, such as cell-, fermentation-, insect-, plant-, fungi- or algae-based alternatives to traditional food ingredients and products. This has brought new challenges in terms of new matrices and/or analytes that need to be tested to assess the quality and safety of these novel foods. In addition, they also challenge traditional analytical approaches, such as the analysis of protein content or quality. However, this challenge presents an opportunity to improve the current methodology and approaches for all food types, not just the novel ones. In addition to the accurate protein determination and evaluation of protein quality, there are other method gaps and concerns related to novel foods, including safety concerns, which require adequate analytical methods for both targeted and non-targeted analysis of various chemical contaminants, residues, toxins, allergens or antinutrients. Consensus on method requirements, development of guidelines and introduction of new standardized methods and approaches are essential to address these challenges and take advantage of these new opportunities in food analysis.

Towards sample-to-solution approaches for food contaminant determination at the point-of-need

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There is a strong drive to get chemical analysis out of the lab, and into the hands of the people that need the (interpretation of) chemical information. These could be consumers that want to know that their food can be safely eaten (e.g. free from allergens or toxins), or inspectors that need to monitor food safety for the government. In all cases, the different steps from an analytical workflow need to be considered from an entirely different perspective when carried out outside the lab. Therefore, different research fields and analytical strategies are combined to achieve such goals, including ambient ionization (portable) mass spectrometry [1,2], lateral flow immunochemistry [3], chemical surface modification for (paper) microfluidics [4] and 3D-printing [5]. In this presentation, Gert Salentijn will elaborate on some recent examples from his work in the field of on-site sample preparation, and portable detection strategies in the context of food safety analysis.

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Plenary Oral Presentations

Long-term targeted and non-targeted monitoring of drinkable water treatment plants in Biscay (Basque Country)

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The presence of emerging contaminants (ECs) in environmental water samples is of increasing concern, especially from pharmaceuticals and personal care products, and by-products of anthropogenic activities such as plasticisers or per/poly-fluorinated substances (PFAS). When some of these compounds are found in untreated and treated drinking waters, the concern requires effective tools to assess the quality of water and the risk to human health.

In order to assess the presence of ECs in untreated and treated drinking waters in Biscay (Basque Country), during two years (5 campaigns per year) we have monitored four drinking water treatment plants (DWTP) and one experimental treatment plant designed to reuse the water from the river. For this, we performed a large volume solid-phase extraction to assure the in situ extraction of more than 30 L of water and implemented a multi-target screening of 280 ECs and a non-target workflow to quantify and identify as many contaminants as possible.

Considering all the DWTPs, we found more than 35 ECs with a frequency above 20% and with concentration levels ranging between 0.1-1000 ng/L. Among the top most frequently found ECs we can highlight BPA, PFOA, 2-hydroxibenzothiazole, valsartan and metformin (freq>45%), showing an average concentration around 10 ng/L. In many of these compounds the average level in untreated and treated waters are very similar suggesting the lack of efficiency in the treatment process at these low levels. Moreover, the temporal pattern was analysed and in some cases, seasonal patterns were observed.

Regarding the non-targeted approach, we were able to identify more than 20 ECs in levels 2A/B belonging to contaminants not included in the targeted analysis (e.g. dodecylsulphate) or metabolites of formerly found ECs.

Finally, though the samples showed negligible cytotoxic and mutagenic levels at enrichment levels of 5, an overall risk assessment analysis of these waters was carried out based on the risk quotients (HQ) for an exposure through ingestion of water (μg/kg/day).

Structural alerts for prioritization of potentially toxic substances in non-target screening

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Text: Feature prioritization is a necessary step in high-resolution mass spectrometry-based non-target screening of environmental matrices. Here, we will discuss a novel prioritization strategy that relies on raw high-resolution mass spectrometry data (MS2) of detected features and the potential presence of structural alerts (molecular substructures that are linked to toxic effects) in MS2's. A data analysis pipeline based on feature selection using frequent pattern mining, followed by a machine learning-based classification algorithm is being developed to predict whether a structural alert is present in detected features, Figure 1. Initially, 7334 compounds included in NORMAN MassBank were screened, based on their SMILES, for the presence of 187 structural alerts using ToxAlerts[1,2] and their 53,610 MS2 spectra were labelled accordingly. Subsequently, either neutral losses (NLs)[3] were computed or fragments were binned to a resolution of m/z 0.01. Finally, a random forest classifier, a single-layered feed- forward neural network and a classifier based on association rules were trained and tested using the two different types of pre-processed MS2 data. Preliminary results (average accuracy of 0.8 and average recall of 0.3) show that further feature reduction as well as the use of alternative strategies such as ensemble learning, might be useful given the large number of predictors and classes.

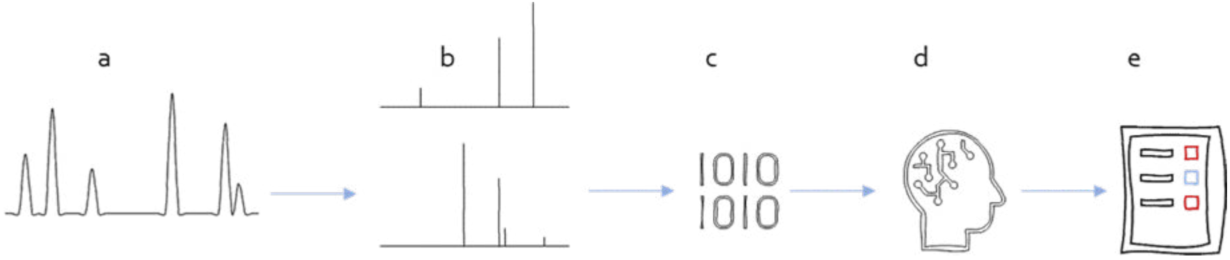

Figure 1 – Intended workflow where (a) an LC-HRMS measurement is conducted, and (b) the corresponding MS2 spectra are
(c) translated into data that can be fed to (d) the machine learning model which predicts (e) the probability that a specific structural alert is present in the feature.

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Two-stage LC/HRMS workflow for identification of effect drivers in passive sampler extracts after their selective recovery by pull-down assay.

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Passive sampling with chromatography coupled to high-resolution mass spectrometry was proved to be a tool for profiling the pollutants in water [1]. Many nontargeted (NT) acquisition methods have been established to detect hundreds of compounds, but the major challenge remains connecting identified chemicals with effect, which stays unexplained for some bioassays up to 98% [2].

In this work, we would like to demonstrate the benefits of pull-down assay application prior to NT analyses for simplification of identifying low abundant effect drivers. In the first stage, full scan data was acquired in positive and negative ESI ionization by hybrid quadrupole/orbital trap HRMS. The dataset was processed for peak picking and differential analysis to remove background, protein artifacts, and nonspecifically adsorbed analytes. The suspect list of the compounds for follow-up MS2 analysis was generated and consequently run in the second step of the workflow.

Combined full scan and parallel reaction monitoring MS^2 data were searched toward online MS^2 spectra libraries, commercial and in-house created mass lists, and ChemSpider database. The number of identified compounds at a level of uncertainty 1-3 on the Schymanski scale [3] was increased by applying mass defect analysis. The involvement of pull-down assay and two-stage LC-HRMS workflow allowed selective identification of relevant environmental pollutants at the ultra-trace level for consequent testing by bioassays.

Acknowledgments
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Determination of real-life environmental and food chemical mixtures in water, fish and milk samples using suspect screening coupled to iterative data-dependent analysis

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Contamination of water, fish and milk by various classes of substances has been widely documented and are matrices of interest in the context of public health, dietary exposure and contamination of the environment. Over the past decade, suspect screening has become a tool for comprehensive analysis of contaminants, providing valuable insights about the presence of emerging contaminants.

The aim of this study was to carry out suspect screening analyses on pooled water and food samples from several European countries. This work was carried out as part of the European PANORAMIX project (Green Deal H2020) coordinated by the Technical University of Denmark (DTU).

Waste-, surface and drinking water from 10 European countries were extracted by solid-phase extraction after filtration of the turbid samples. Fish and cow milks samples from 7 to 8 European countries were prepared by liquid-liquid extraction including a delipidation step. The extracts were analysed by LC-HRMS (Q Exactive) using iterative data-dependent analysis on pools, followed by the injection of individual samples in Full MS mode. Data were processed by the Galaxy Workflow4Metabolomics tool after conversion to mzML universal format. The detected features were prioritized, according to a combination of matching and scoring criteria, leading to a list of detected markers associated with identification confidence levels ranging from 3 to 1 for each considered matrix, according to the Schymanski scale. For those markers for which appropriate standards were available, semi-quantitative concentrations could be estimated.

A wide range of contaminants was identified, with pharmaceuticals being one of the most abundant chemical classes, along with pesticides and cosmetics, mostly in water. Some candidates were found in common in several matrices studied. Unsurprisingly, extracts from wastewater showed the highest number of markers with the highest estimated concentrations. Several parent-metabolite pairs have also been tentatively detected supporting the relevance of considering the transformation of chemicals (metabolism and environmental degradation). However, currently existing databases still suffer a lack of spectra related to transformation products, so that this comprehensive chemical annotation process still faces significant limitations.

Identification of emerging organic contaminants in greywater emitted from ships by a comprehensive LC-HRMS target and suspect screening approach.

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Shipping has long been recognized as an energy-efficient mode of transport medium for moving freight. However, with the increase of maritime traffic, large proportions of wastewater are continuously being released into the marine environment, posing potential threats for vulnerable marine ecosystems. Among the different types of wastewaters released from ships, greywaters account for the larger proportions to the total volume of wastewater produced [1,2] Since these waters are more concentrated than land-based wastewaters, it is of outmost importance to perform a comprehensive chemical characterization of greywaters emitted from ships to fully assess the potential environmental and human health risks associated with their discharge to the marine environment. The aim of this study is to use the benefits of HRMS analysis, by combining wide scope target (>700 compounds) and suspect screening methods, using an Orbitrap Exploris^{TM} 120 instrument, for the holistic characterization of greywater. Samples were provided by BSAG, who collected greywaters from cargo ships docked at Helsinki port in Finland, as part of the Horizon 2020 EMERGE project (Grant agreement ID: 874990). For target analysis, an established method, that included the exact mass of precursor and fragment ions and the retention time, was used [3]. Suspect screening was conducted by the software Compound discoverer 3.1 (Thermo Fisher Scientific), using the software-linked databases, mzCloud, mzVault and Chemspider, for compound identification in one single analytical platform. With this holistic approach, 119 compounds were tentatively identified, 87 by target and 32 additional substances by suspect screening. Furthermore, 44 out of the 87 target compounds could be further validated and quantified by analytical standards. The substances identified covered several chemical classes, including pharmaceuticals, personal care products, perfluoroalkyl substances, plasticizers, and natural products.

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Trend analysis of non-target screening features in surface water for safe drinking water production

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In the Netherlands, surface water from the river Rhine and river Meuse is a crucial source for drinking water production. However, the chemical complexity and large fluctuations in surface water quality prove to be challenging to ensure safe drinking water in the future. Consequently, comprehensive monitoring is conducted on these drinking water sources to assess the occurrence of diverse substances, like micropollutants and emerging contaminants. Starting from 2019, the Water Laboratory in Haarlem and Aqualab Zuid in Werkendam have been routinely employing LC-QTOF non-target screening (NTS) analyses to examine the presence of unidentified compounds (features) at drinking water intake locations in the Rhine and the Meuse. In practice, the data analysis and the data interpretation has proven to be challenging, since NTS data is abstract and extensive, hindering its translation into practical applications. Cross-laboratory, we have extended a workflow that facilitates NTS data processing and retrospective data analysis [1,2]. Due to the considerable volume of information generated through data processing, prioritizing features becomes crucial. One way to primarily prioritize features is to apply trend analysis, to reveal patterns and changes in the concentration or occurrence of the various unknown substances present at the drinking water sources. Features showing significant trends, may be given higher priority as they could indicate potential risks or opportunities for further research. In addition, we explore the usage of other feature prioritization tools such as exploring the MS2 spectra for possible structural alerts.
In this presentation we present a workflow for the analysis of 4 years of routine NTS data using the open-source tool patRoon [2] in combination with extensive trend analysis techniques to prioritize unknown compounds at drinking water intake locations from the Rhine and the Meuse. This approach yields a comprehensive list of significant features and their variations over time. Moreover, this modular workflow easily integrates with other prioritization techniques for water quality monitoring with NTS and is extendable to work cross-laboratory.

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Decoding the human exposome: monitorization of chemicals of emerging concern and their phase II metabolites in urine by target, suspect and non-target screening

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In the context of decoding the human exposome, the analysis of urine has great potential since it is the main biofluid for excreting contaminants of emerging concern (CECs) and their metabolites from our organism. Considering the large number of CECs present in our daily lives, high-resolution mass spectrometry (HRMS) is a promising tool to evaluate our exposure to them, since it allows the detection of a wide range of compounds. Additionally, HRMS offers the opportunity to look for CECs and their metabolites using suspect and non-target screening (SNTS) [1]. In the present work, a diversity of 200 CECs was monitored using target analysis in 150 urine samples collected among students and workers from the University of the Basque Country (Spain). Furthermore, the analysed chemical space was widened by SNTS including more than 8000 CECs obtained from NORMAN and their respective phase II metabolites (~ 8000) simulated by BioTransformer 3.0. The urine samples were treated with and without enzymatic hydrolysis using β-glucuronidase/ arylsulfatase, extracted with Oasis HLB cartridges, and analysed by UHPLC-qOrbitrap. Regarding the results, diverse CECs were quantified in the samples by target analysis (Figure 1a). In SNTS, 348 exogenous suspects were annotated avoiding false positives since a list of more than 2500 endogenous substances was employed (Figure 1b). In addition, phase II metabolites were also screened (Figure 1c).

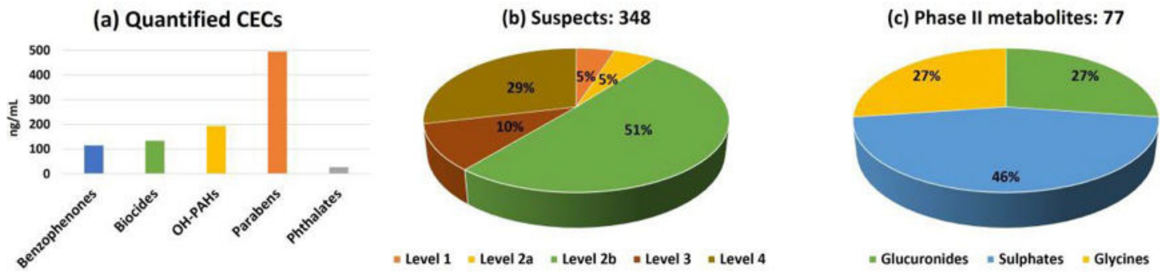
(a) Quantified CECs
(b) Suspects: 348
(c) Phase II metabolites: 77

Figure 1. (a) Target analysis results, (b) annotated suspects by SNTS, and (c) screened metabolites by SNTS.

Lastly, Principal Component Analysis (PCA) showed that creatinine content was positively correlated with age and higher concentrations of the CECs. Moreover, women differed from men especially because of benzophenones, which are linked to the higher usage of cosmetic and personal care products reported in the personal questionnaires.

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Using LCMS Analysis and Suspect and Non-target Screening to Assess the Efficiency of Hybrid Electrodialysis-Forward Osmosis (ED-FO) in Water Reuse

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Anaerobic digestion (AD) is a well-established and popular technology used in the treatment of organic waste produced in water treatment plants. During the anaerobic digestion, microorganisms break down organic materials in an oxygen-free environment, converting organic matter into biogas (i.e., methane) and a nutrient-rich residual sludge known as digestate. The anaerobic digestate, and particularly its liquid fraction, is commonly used as soil amendment as a substitute for synthetic fertilizers. This application has raised concerns due to the presence of heavy metals, pathogenic organisms, and xenobiotic contaminants in the digestate which could pose a risk to human, animals, and ecological systems. In this work, we aim to introduce and test the efficiency of an innovative hybrid electrodialysis-forward osmosis (ED-FO) process designed to safely extract valuable nutrient ions and clean water from the anaerobic effluent. The major objectives of this work are to demonstrate the feasibility of the proposed treatment process via well-controlled bench-scale experimental setups and to evaluate the removal of xenobiotics, including contaminants of emerging concern. The AD effluent sludge (raw liquid digestate) was collected from the Gresham Wastewater Treatment Plant in Oregon, US. Samples were subjected to a pre-treatment process for solid-liquid separation including centrifugation, flocculation, sedimentation, and filtration. After the pre- treatment, the recovered liquid was treated by ED followed by the FO process. Three replicates per sample type (raw, pretreated, and post-treated) were collected and processed for analysis. Organic chemicals were analyzed via ultra-high performance liquid chromatography (UPLC) high-resolution mass spectrometry (HRMS). Prior to LCMS analysis, samples were filtered through a 0.45 µm nylon membrane and preconcentrated 500 times using a solid phase extraction (SPE) automated system. Data was processed using SCIEX OS and MS-DIAL[1] software. From the 260 annotated compounds using spectral libraries, 230 (88.5%) were significantly reduced after the ED-FO process. Toxicity of annotated compounds was subsequently evaluated *in silico* using a Hazard Cheminformatics Module[2] developed by the EPA, US. Our results show the efficiency of this treatment technology as an innovative application in water reuse processes.

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Data science enhanced non-targeted analysis for characterization of exposome: move beyond structural elucidation

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Resolving the human and environmental exposome is an extremely challenging task due to the complexity of their chemical space (e.g. the number of potential structures). Liquid chromatography coupled with high resolution mass spectrometry (LC-HRMS) is one of the most dominant and comprehensive techniques for non-targeted analysis (NTA). This combination has been widely employed for tackling the complexity of exposome. The NTA approaches, even though powerful, are limited to the identified portion of samples. For vast majority of the chemical constituents in the samples, no inferences regarding their toxicity and fate can be made as they remain structurally unknown (i.e. unidentified). Here we report the development of a set of data science and computational mass spectrometry tools that use the acquired MS/MS information to infer about the environmental fate and biological activity of structurally unknown chemicals. Both toxicity categories and the ionization efficiencies of the unknown chemicals were predicted based on the MS2 spectra. This information resulted in a quick and relatively accurate estimation of the risk for the chromatographic features, without the chemical identification bottleneck. Additionally, using the MS/MS information a model was built to predict the retention behavior of structurally unknown chemicals. This enabled the alignment of LC-HRMS chromatograms, thus providing the means to perform trend analysis across different chromatograms acquired using different methods. Finally, the network analysis enabled the clustering of the chemical structures that could potentially go through similar reaction mechanisms in the environment, thus providing insights into transformation products of structurally known and unknown chemicals.

PERK: A Cutting-Edge R-Based Tool for Enhanced Environmental Risk Assessment of Active Pharmaceutical Ingredients in Wastewater

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Abstract:

The accurate assessment of environmental risk posed by active pharmaceutical ingredients (APIs) requires sophisticated modelling approaches to predict their concentrations in the environment. This study uses PERK[1], an advanced R-based open-source software tool designed to streamline and enhance environmental risk assessment (ERA) by automating modelling and reporting of predicted environmental concentrations (PEC) and risk quotients (RQ) for a comprehensive range of APIs.

PERK stands out as a cutting-edge solution, leveraging the power of R and incorporating innovative spatiotemporal data filters for unparalleled accuracy in environmental research. The platform features an intuitive interface as an interactive R-Shiny online application with a state-of-the-art dashboard architecture, empowering users to effortlessly analyse and visualize pharmaceutical concentrations in various environmental matrices.

In addition to prediction, PERK excels in visual analytics, enabling the visualization of removal efficiency, prediction accuracy, and measured concentration, load, and population-normalized load based on user inputs. This comprehensive visual representation empowers researchers to gain deeper insights into API behaviour in wastewater systems.

Using high spatial resolution prescription data collected at wastewater catchment level[2], PERK efficiently extracts and processes API prescription data. Its distinctive database with $F_{excreta}$ values ensures robust and reliable API concentration modelling.

Application of PERK in a study involving wastewater samples from major South West England wastewater treatment plants[3] demonstrated its ability to deliver accurate predictions for 12 pharmaceuticals and acceptably accurate results for 24 out of 28 pharmaceuticals in prescription-only medicines. Elevated $PEC_{effluent}$ levels for pharmaceuticals with high consumption and low removal rates were identified, raising environmental concerns.

In conclusion, PERK represents an innovation in ERA, providing a potent, user-friendly tool that enhances the accuracy and efficiency of API concentration modelling. Its unique features and analytical capabilities, combined with advanced visual analytics, support informed decision-making and sustainable environmental management on multiple temporal and geographical scales.

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Elimination resistant: Modelling multi-compartment toxicokinetics of thiacloprid in *Gammarus pulex* using monitoring data, bioconcentration and receptor binding assays

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Neonicotinoid insecticides act as agonists of the nicotinic acetylcholine receptors (nAChRs) and induce symptoms of neurotoxicity. In aquatic invertebrates, delayed toxicity is a common phenomenon for neonicotinoids. Monitoring of thiacloprid in surface water and aquatic organisms during the application season revealed continuous internal body burden even if no pesticides were found in the water.[1] Lab studies with amphipods confirmed the incomplete elimination suggesting multi-compartment toxicokinetics. We hypothesised that irreversible receptor binding might be the mechanistic link to the observed elimination resistance. To test this hypothesis, toxicokinetic experiments with *Gammarus pulex* were complemented with studies on the binding of thiacloprid to nAChRs using *in vitro* and *in vivo* receptor binding assays.[2] Using an online-solid phase extraction system coupled to LC-HRMS/MS allowed the quantification of thiacloprid down to the receptor-bound thiacloprid fractions recovered from gammarid membrane protein extracts. Based on the receptor binding assays, irreversible binding to the nAChRs was demonstrated to be the major cause of elimination resistance. Accordingly, a toxicokinetic-receptor model was developed that predicted thiacloprid concentrations in gammarids successfully. The model consists of a membrane protein (i.e., nAChR) compartment and a structure compartment (Figure 1). In the future, it can be adjusted to assess other receptor binding compounds and mechanisms, and can be implemented into the environmental risk assessment. In this way, long-term toxic effects following the irreversible binding to receptors can be addressed and regulatory solutions explored.

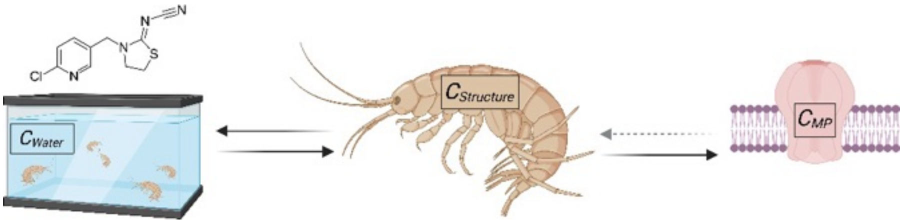

Figure 1: Schematic illustration of the toxicokinetic-receptor model. Arrows indicate contaminant fluxes. *C* = Concentration, *MP* = Membrane protein

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Prediction of Acute Fish Toxicity using Molecular Fingerprints and Machine Learning

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The human exposome encompasses a vast number of chemicals, estimated to range from 350,000 to 800,000. However, our understanding of the fate and behavior of these chemicals is limited. Conducting experimental studies on such a large scale is unfeasible, which is why modeling approaches are being utilized.

One commonly used modeling approach is Quantitative Structure-Activity Relationships (QSAR) models, which rely on molecular descriptors. These descriptors are mathematical representations of a molecule's properties. However, a drawback of this approach is that the calculation of molecular descriptors is not stable. As a result, predictions made by the model for the same molecule can vary between calculations, leading to unreliable outcomes. Therefore, it is important to explore alternative modeling sources. Thus, alternative modeling sources should be explored. Molecular fingerprints are an alternative to molecular descriptors that represents the structure of the molecule, rather than its properties, and can be directly associated with the target activity or property.

In this study, we present a random forest regression model for predicting acute fish toxicity using molecular fingerprints. We collected relevant variables for toxicity prediction from different types of fingerprints and combined them into a master fingerprint. The final model, with model scores of ~0.9 for the training set and ~0.7 for the test set, demonstrates that our regression model results are comparable to similar descriptor-based models [1]. With molecular fingerprints advantages in terms of consistency and interpretability, they provide a promising alternative to molecular descriptors for assessing chemical behavior. Furthermore, we have developed an algorithm to optimize the selection of important variables for the prediction of specific properties. This algorithm makes developed approach universal and applicable to different case studies.

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Agglomeration behaviour and fate of food-grade titanium dioxide in human gastrointestinal digestion and in the lysosomal environment

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The fate of nanomaterials during human digestion is still poorly understood and available evidence indicates that the interactions with the gastrointestinal (GI) environment may alter the physicochemical properties of ingested particulate materials and affect the intestinal uptake of the particles and their toxicological properties. New Approach Methodologies (NAMs) have enormous potential to improve mechanistic understanding of toxicokinetic and toxicodynamic processes at the nanoscale by focusing on human relevant models. Integrated Approaches to Testing and Assessment (IATAs) are then used for the integration of human, animal and NAMs-derived evidence. Focus is on particles up to approximately 250 nm since available evidence suggests that specific cellular uptake mechanisms exist for particles within such size range. Food-grade titanium dioxide (E 171) is a widely used food additive which, following the recent safety re-assessment of the European Food Safety Authority (EFSA), has been banned for such use in the European Union (EU). Other bodies worldwide have subsequently assessed the risk of titania as food additive coming to different conclusions, also on the basis of simulated GI digestion studies in which light scattering techniques were used to get information about the size of the TiO_2 particles in suspension. However, these techniques are not suited for screening the presence of small particles or characterising particle size distributions (PSDs) of polydisperse materials and, as such, are not accepted for regulatory safety assessment in the EU.

In the present study, we addressed the knowledge gaps and the uncertainties regarding the agglomeration behaviour and fate of food-grade titanium dioxide in human GI digestion. A representative sample of E 171 was submitted to a thorough physicochemical characterization using a state-of-the-art multi-technique approach and its GI fate was studied by applying the in vitro GI digestion approach laid down in the EFSA guidance on risk assessment of nanomaterials, in both fasted and fed conditions. In addition, real food samples containing E 171 were also studied. Single particle ICP-MS was the key technique in the present study to characterize the agglomeration behaviour of E 171 and the obtained results showed that light scattering techniques used in earlier studies delivered biased results. We demonstrated that, after simulated GID, in the small intestine E 171 at concentrations reflecting human exposure is well dispersed, with PSDs having ≥70% of the particles <250 nm. In addition, since TiO_2 particles were found to be resistant to GI dissolution, stability in lysosomal fluid was investigated. Biopersistence of the material in lysosomal fluid highlighted its potential for bioaccumulation.

In-vitro digestion approach for assessing the bioaccessibility of pharmaceuticals in seafood

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Pharmaceuticals have been ubiquitously detected in the environment and showed potential to bioaccumulate in aquatic organisms (Álvarez-Muñoz et al., 2018), representing a potential risk for human health due to dietary exposure. However, the level of a pharmaceutical detected in seafood does not reflect the quantity that will be released from food during the gastro-intestinal digestion, becoming available for absorption in the intestinal lumen. *In-vitro* digestion methodologies are considered a suitable tool to simulate the human digestion, allowing to evaluate the bioaccessibility of contaminants in seafood (Trujillo-Rodríguez et al., 2020). In this context, an *in-vitro* digestion approach was applied for accessing the bioaccessibility of three selected pharmaceuticals (ofloxacin, citalopram and venlafaxine) in raw and cooked mussels. The simultaneous presence of other emerging contaminants (e.g., microplastics) may alter the food concentration of pharmaceuticals and, therefore it was also considered in this study by evaluating the effect of the simultaneous presence of microplastics (PE and PS) in the bioaccessibility of the selected pharmaceuticals.

In raw mussels, the bioaccessibility of pharmaceuticals varied between 97 and 123%, while in cooked mussels this was lower (71-91%). The presence of microplastics generally led to a small decrease in the bioaccessibility of pharmaceuticals, except in cooked mussels where the presence of PS microplastics showed a small increase in the bioaccessibility of citalopram and venlafaxine. This study gives new insights on the bioaccessibility of pharmaceuticals that can help to obtain a more accurate assessment of health risks due to intake of pharmaceuticals-contaminated seafood.

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Effect-directed analysis in the environment-food-human continuum to identify chemicals with Transthyretin binding properties

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Effect-directed analysis (EDA) is an innovative approach used to identify and evaluate the potential risk posed by complex mixtures of chemicals. It combines state of the art analytical techniques with *in vitro* bioassay testing to pinpoint the specific components responsible for the observed toxic effects[1,2]. EDA approaches typically include: testing the biological activity of the original extract, followed by fractionation, repeated bioassay testing and chemical analysis of the pre-selected samples. In this study, a high-throughput EDA approach was used to assess the potential of chemical mixtures present in water, fish, cow and human milk, and human serum samples to interfere with thyroid hormone (TH) serum protein binding, which is a key molecular initiating event related to disruption of the TH system. Environmental and drinking water, fish, cow and human milk, as well as human adult and cord blood samples from several countries were extracted using different techniques. The extracts were processed accordingly for assessment of their capacity to compete with TH for binding the TH distributor protein transthyretin (TTR)[3]. Samples showing higher TTR-binding effects were subjected to high throughput EDA[1]. Briefly, 13 selected extracts and their matching procedural blanks were fractionated by high performance liquid chromatography (HPLC) coupled to a fraction collector (FractioMate™). Non- targeted chemical profiling of the extracts was performed using the same chromatographic conditions coupled to a qTOF mass spectrometer. Annotation of the detected features (suspect screening data processing) was performed using the Galaxy Workflow4Metabolomics tool. The activity of the fractionated extracts was assessed using the same TTR-binding assay.

Several active fractions were detected in all samples considering an 20% effect threshold. No common bioactive fractions could be seen between different sample types indicating that different chemicals are responsible for the mixture effects observed in the different matrices. Several analogies could be seen in human serum samples. Common active fractions were also found between organic and conventional cow milk. The alignment of the bioassay results with the corresponding mass spectrometry-chromatogram and corresponding feature identification will be presented.

The presented high throughput EDA covered samples representative of the environment-food-human continuum, showing that activity drivers might differ between sample types, but can be conserved within the same source.

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Pull-down approach for the identification of endocrine-disrupting compounds in complex mixtures

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The environment can contain a wide range of micropollutants in highly complex mixtures, which may cause adverse effects in exposed biota or humans. Endocrine disrupting (ED) potential through various modes of action has been frequently detected by bioassays in environmental samples. Despite the long list of analytes measured in recent studies, only a relatively small portion of the observed effects can often be explained by detected chemicals, and the causative chemicals remain largely unknown. Approaches for prioritizing and identifying the drivers of these effects include mixture and QSAR modeling and effect-directed analyses.

This contribution introduces a novel pull-down approach for identifying ED compounds in complex mixtures, which utilizes engineered recombinant target proteins as specific baits for their ligands. The target proteins for the endocrine-disrupting compounds include some crucial nuclear receptors or hormonal transporters. We will present the development and optimization of a method enabling the separation and identification of compounds interfering with hormonal transport by binding to transthyretin (TTR), a serum transporter protein of thyroid hormones. TTR-binding inhibition has been associated with many adverse effects in different species. This method uses an in-house purified recombinant TTR protein as bait in the pull-down assay. We applied the optimized TTR pull-down method in a case study focused on treated wastewater (WW) and impacted small stream. Eluted ligands are analyzed in the TTR inhibition bioassay and non-target screening workflow based on HPLC-HRMS, which enables differential analysis of pull-down assay eluates and controls. This approach largely reduces the very complex mixtures to the specifically bound ligands. We have identified several known ligands as well as a set of novel ligands for TTR. In case of standard availability, these compounds were verified and quantified in the original samples, and their effects and potencies were determined through bioassays. The mass balance calculation showed high explicability of the observed effects by the identified compounds. The presented method is a promising tool for identifying bioactive compounds and prioritizing risk drivers from complex exposure mixtures.

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The contribution of PFAS to thyroid hormone-displacing activity in Dutch waters: A comparison between two in vitro bioassays with chemical analysis.

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Per- and polyfluoroalkyl substances (PFAS) are a group of xenobiotics that are widely distributed throughout the aquatic environment.[1] Many PFAS are possible thyroid hormone (TH) system disrupting compounds, because they have the capacity to -amongst other- inhibit the TH thyroxine (T_4) from binding to its transport protein transthyretin (TTR).[2] This study investigated the occurrence of TH-displacing activity in the Dutch (drinking)water cycle, and more specifically, the contribution of PFAS to this effect. Over one year of monitoring data of 29 PFAS (linear and branched) showed the continuous presence of PFAS in drinking water (DW) and surface water (SW) sources used for their production. Two bioassays that measure TH-displacement –one (FITC-T_4) was a binding bioassay and the other (TTR-TRβ-CALUX) a cell-based reporter gene assay- were compared using spiked samples and relative potency factors (RPFs) for TH-displacement. They were found to be suitable for measuring TH-displacing activity in water samples. As a third aim, a field study was performed in the Dutch water cycle that was comprised of samples from DW, SW, PFAS hotspots, and 2 wastewater treatment plants. Predicted TH-displacing activity of up to 20 target PFAS revealed that analyzed PFAS only explained ≤4.1 % of activity measured in water extracts by both bioassays. This indicated that as yet unknown compounds contribute to the majority of the measured TH-displacing activity. Current work focuses on unraveling the identity of these compounds by effect-directed analysis (EDA) to explain the currently missing gap of TH-displacing activity measured in water samples.

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Optimizing analytical approaches in high-throughput effect-directed analysis (HT-EDA) for improved toxicity driver identification

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Effect-directed analysis (EDA) has established itself as one of the most promising tools for unravelling the complexities of environmental samples and elucidating toxicity drivers. Although the progress in the field of non-targeted analysis has improved the success of recent EDA studies, the time and labour-intensive nature of EDA prevent its routine applications. The development of new high-throughput (HT) systems for sample fractionation, as well as advances in high-throughput toxicity assessment, have moved EDA forward. Microfractionation of contaminant mixtures in 96-well plates using new fractionation systems allow the implementation of faster and more efficient EDA methods, especially when these plates can be tested with bioassays in a high-throughput manner[1].

Still, the risk drivers might be overlooked (i) if the chemical analysis is limited to single ionization preferences which confines the detectable chemical space, or (ii) if the spectral quality is not good enough to allow annotation with a high confidence level. To increase the success of identifying toxicity drivers in HT-EDA, our study focuses on the optimization of analytical approaches using androgenicity as selected endpoint. This new workflow includes:

- A new strategy in which data acquisition and processing will not be limited to the raw sample but the data will be acquired also from the toxic fractions.
- Optimized chromatographic, ionization, and MS conditions for the analysis of toxic fractions (e.g., ESI vs APCI, adapted chromatographic gradient, scan time/speed…).
- Non-target screening workflows to prioritize potential toxic compounds (e.g., differences in target ions/adducts), including endpoint-specific suspect lists (for androgenic agonists in this study), and spectral alarms to prioritize structures potentially related to these endpoints[2].

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Effect-directed analysis for multiple endpoints of bioactive compounds in the (drinking) water cycle

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Surface waters (SW), such as the rivers Rhine and Meuse are important sources for the drinking water (DW) supply in Europe. DW companies therefore intensively investigate these sources for the presence of known and emerging contaminants and their fate during treatment processes. The Water Laboratory uses e.g. a panel of reporter gene bioassays with endpoints relevant to human and ecosystem health to this aim. In cases when elevated activities in bioassays of unknown cause are observed, a high resolution Effect-Directed Analysis (EDA) platform is applied to identify the causative compounds (1). It combines LC-QToF-MS with parallel bioassay detection according to a workflow we developed for use in e.g. a legislative, governmental or routine monitoring context (2).

This presentation discusses examples of research performed with the panel bioassays and consecutive identification of bioactive compounds with EDA. One study investigated the presence of activities for 10 endpoints in WWTP effluent, SW and DW of 4 DW companies in the Rhine and Meuse catchments. Eight out of 10 endpoints were detected in the samples; most of which were completely removed in DW treatment. EDA was successful in identifying bioactive compounds, although not all peaks could be resolved. Examples of identified compounds are e.g. azole fungicides as compounds causing anti-androgenic activity and steroid hormones (e.g. DHT, (medroxy-)progesterone) causing hormone-like effects.

Apart from the EDA, the recorded targeted MS and bioassays data were analyzed with multivariate analysis (MVA). A clear distinction appeared between the Rhine and Meuse catchments, reflected by separate clusters of Rhine versus Meuse samples. DW samples often clustered far away from their own source samples, indicating that treatment is even more dominant in determining the water quality of DW than the source used. Correlations between bioassay responses and target compounds in MVA were used as possible confirmation of causative compounds delivered by EDA.

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Novel Effect-Direct Analysis (EDA) approach to assess endocrine activity of food mixtures

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Foods are complex mixtures comprising thousands of chemicals, with many of them being structurally uncharacterized and among which, some could present endocrine properties. Assessment of endocrine activity in food and food-related items has taken considerable attention at regulatory and scientific level. From the toxicological perspective, measuring endocrine activity cannot inform on endocrine disruption and/or health risks without sufficient knowledge on the nature of the responsible factors. Effect-direct analysis (EDA) have been promoted for their potential to detect bioactive substances providing information on interactions in complex mixtures. However, reliability of the EDA in complex mixtures is challenging and accurate testing strategies are needed including both, suitable extraction methods to avoid data (mis)interpretation and capabilities to identify the bioactive compound(s) to assess risk assessment.

The topics addressed in this study includes: 1) importance of the extraction method used for EDA; 2) fit-for-purpose battery of in vitro bioassays to assess estrogen and androgen effect and 2) chemical identification of bioactive compounds and concordance analysis. Breakthrough methodology to improve the battery of endocrine bioassays was developed using High Performance Thin-Layer Chromatography (HPTLC) coupled to the Estrogen and Androgen Screen Yeast assay (p-YES and p-YAS). Finally, the HPTLC method was integrated to Liquid Chromatography Mass-Spectrometry (LC-MS) for bioactive compound(s) identification.

The novel EDA battery will be presented as well as a case study (soy isolate) to illustrate the performance of the HPTLC approach to identify bioactive compounds in mixtures as previously shown (1). Altogether, the data generated suggest that the integration of novel tools to the EDA is a powerful approach to better characterize the endocrine activity of complex mixtures.

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Combatting cannibalism: early detection of processed animal proteins in poultry feed

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As a result of the partial lifting of the feed ban in 2021 (Regulation (EU) 2021/1372), processed animal proteins (PAPs) and hydrolysed proteins (HPs) derived from poultry, pigs and insects are re-authorized in feed materials. However, the ban on cannibalism, and the restriction on the use of PAPs derived from cattle, stay in place. To ensure compliance with these restrictions, food safety authorities perform controls in feed-producing factories. Currently, the authorities only send limited numbers of samples to official control laboratories for analysis, as those sensitive, robust and reliable methods are time-consuming and expensive. Additionally, food safety control would benefit from early identification of potentially high-risk sample streams, with a higher chance of intra-species cross-contamination of the produced feed. To enable early identification, and thereby stimulating risk-based monitoring, we have developed easy-to-use, at-location applicable Loop-mediated amplification (LAMP) assays for the detection of poultry (chicken and turkey, ducks and geese as defined by the European Commission[1]) in highly processed feed materials. Performance of the developed assays was validated in (fresh) meat mixtures spiked with poultry meat, and highly processed feed materials spiked with poultry PAPs/HPs. Although currently no legal limit is set for the presence of PAPs/HPs in feed, here 0.1% admixture has been defined as limit, as this is sufficient for monitoring cannibalism and authenticity[2]. Results show a detection limit of 0.01% in spiked chicken meat, whereas, in severely processed meat (and bone) meal 1% could be reached, and in feed materials with unknown processing conditions 0.05% spiked PAPs were still detected. Comparison of LAMP with the EURL-AP recommended qPCR method[3] showed that LAMP performs on par with qPCR in (fresh) meat mixtures. However, in highly processed feed materials qPCR was found to be more sensitive: 0.1% spiked PAPs were still detected, compared to 1% with LAMP. To show true at-location applicability, the developed LAMP assays were combined with easy-to-use DNA extraction and amplification devices[4]. In conclusion, the developed at-location methods can be used for the identification of high risk areas in feed manufacturing factories, thereby facilitating risk-based monitoring required to ensure compliance with the prohibition of intra-species recycling in feed materials.

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Three sampling strategies for microplastics analysis in beaches: A critical comparison

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Microplastics (MPs) have been defined as small pieces of plastic less than five millimetres in length that can be harmful to our oceans and aquatic life. Today, they are present in all environmental compartments, including oceans. The less dense MPs present in the oceans tend to accumulate on beaches. Unfortunately, there is no generally accepted field method for collecting sand in order to quantify and characterise the MPs present in it.

We have tested 3 methods for collecting sand samples on beaches. The first is a judgemental method, whereby 4 operators collect MPs subjectively for 2 hours along a 100 m transect of the beach. The 1-5 mm fraction is transported to the lab in glass containers. The second has been proposed by the Marine Strategy Framework Directive (MSFD). Briefly, at each sampling point the top 1 cm of sand is collected within a 50x50 cm quadrant and sieved in situ to retain the 1-5 mm fraction to be transported to the lab in glass containers. The third has been proposed by our research group. 5 subsamples are collected in each 50x50 quadrant using metal cans of known dimensions, which are transported to the lab for further handling.

These 3 methods were carried out at two different beaches of the Bay of Biscay in November 2022. When necessary, the MPs were separated from the matrix by flotation in a concentrated NaI solution before filtration through a 1 mm metallic filter. All the MPs obtained were individually weighted, classified (colour and shape), and characterised by FT-IR and Raman spectroscopies (PE, PP, PS, others). The results were reported in terms of concentration (mass of MPs/m^3) and percentage according to chemical composition, colour and shape.

The judgmental method produced biased results, apparently, due to the tendency of the samplers to collect white pellets rather than other MPs. Similar results were obtained with the other two methods in terms of the distribution of MPs according to shape and colour. However, the MP concentration was lower with our method than with the MSFD one. Our method considerably simplifies the work to be done on the beach, but the processing of the samples in the laboratory is harder and more time-consuming.

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Towards rapid on-site detection of atropine in cereals

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The number of reported food contamination cases with tropane alkaloids (TAs) is growing worldwide. The current identification tools for these contaminants are time-consuming, laboratory-bound, and expensive. Consequently, screening of food commodities on TA contamination is only feasible in countries with government agencies and companies that have enough financial and human resources. Therefore, there is a pressing need for inexpensive and on-site applicable screening methods for TA contaminations. From the 200 TAs described in literature, the two regulated by the European union are scopolamine and atropine. All TAs are weak bases, making the polarity of these compounds pH-depend. In this work, we developed a prototype immunosensor in which this characteristic was employed. The prototype consists of a user-friendly and inexpensive TA-generic sample preparation combined with an atropine-specific indirect competitive lateral flow immunoassay (icLFIA) for analysis. TAs were extracted from the buckwheat cereals by a mixture of base aqueous and apolar organic solution, to minimize matrix co-extraction. The TAs were subsequently concentrated with a paper-immobilized liquid phase micro extraction (PIPME), i.e. acidic water. The difference in capillary pressure of the acidic aqueous and apolar organic solution for the paper ensured phase separation. This acidic paper was integrated in a 3D-printed extraction frame, which can be inserted in a corresponding 3D-printed elution holder. After elution, the holder can be directly coupled to the icLFIA casing to transfer the concentrated atropine directly towards the icLFIA for analysis, increasing user-friendliness. Digital read-outs of the icLFIA, by employing a hand-held reader, enabled semi-quantification of the atropine concentration (IC_{50} = 0.56 ng mL^{-1}). The applicability of the prototype immunosensor was demonstrated by successfully analyzing blank and spiked buckwheat cereal samples at two different target levels (5 and 10 ng mL^{-1}). Therefore, the prototype immunosensor has the potential to aid in the on-site detection of TAs by non-experts. In addition, the sample preparation is suitable for other compounds with a pH dependent polarity.

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Experimental and computational optimization of polymer inclusion membrane technique for mineral extraction in seawater

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There is a significant increase in demand for minerals as they play a huge role in the economy. This study reports on the experimental and computational development and application of polymer inclusion membrane (PIMs) for the extraction of mineral salts in seawater. The PIMs were synthesized by incorporating base polymer such as cellulose triacetate (CTA), thenoyltrifluoroacetone (TTA) and trioctylphosphine oxide (TOPO) as carriers and o-nitrophenyloctyl ether (NPOE) as plasticizer. The optimization was conducted by varying the composition of the membrane, pH, concentration of the receiver solution and extraction time due to their influence on the extraction efficiency of the targeted analytes. The synthesized membrane was composed of 60 wt.% (TTA+TOPO) carriers, 30 wt.% (CTA) base and 10 wt.% (NPOE) plasticizer (1:1) ratio. The selectivity of the optimised method in seawater extraction was as follows: $Mg^{2+} > Ca^{2+} > Li^{+}$. The other targeted competing species such as sodium and potassium were not detected by the inductively coupled plasma optic emission spectroscopy (ICP-OES). The results were supported by computation studies that showed that with TOPO as carrier, the selectivity was as follows: $Mg^{2+} > Ca^{2+} > Li^{+} > Na^{+} > K^{+}$ while TTA showed the following selectivity: $Mg^{2+} > Ca^{2+} > Li^{+} \sim Na^{+} > K^{+}$

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Demonstrating the reliability of metabolomics-based chemical grouping: Towards acceptable practice

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Metabolomics has reached a critical point in determining its value to regulatory toxicology. Building on 20 years of research, the first metabolomics data to support grouping/read-across was recently submitted to ECHA, metabolomics best-practices for various applications including grouping were published in Nature Communications, and the OECD Omics Reporting Framework has been developed. Given these and other examples of its increasing relevance to chemical safety regulations, an assessment of the reproducibility of metabolomics in the context of chemical grouping is required. The aim of the Cefic-funded MATCHING study (MetAbolomics ring-Trial for CHemical groupING) is to determine whether this technology can demonstrate high reproducibility in grouping, and hence high reliability, or whether refinements in analytical or data analysis practices are needed. Through this fully-blinded evaluation, the second aim is to propose 'acceptable practice' for metabolomics-based grouping. The international consortium comprises seven industrial, government and academic metabolomics ring-trial partners, BASF SE, and the European Chemicals Agency (ECHA) as an independent advisor. First, 8 substances were selected for the trial, and all ring-trial partners were fully-blinded to their identities, modes of action, and the number of categories. Plasma samples for the ring-trial were then derived from 28-day rodent tests (8 substances, each 'low' and 'high' dose, plus vehicle controls), aliquoted, and distributed to partners. Each metabolomics laboratory then applied their preferred LC-MS metabolomics workflows to acquire, process and statistically analyse the plasma samples. This included attempting to group the 8 substances into categories based on their metabolomics signatures, and then reporting their findings to ECHA to ensure the blind conditions of the trial were respected. Six ring-trial partners have reported, and 5 have discovered the identical grouping of the 8 substances into 3 unique categories (remaining partner sees similar trends but had more analytical variability). In conclusion, the findings from the MATCHING study have demonstrated high reliability of metabolomics-based chemical grouping.

GC×GC for the determination of fatty acids in cow colostrum during the first days of lactation

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Bovine colostrum is an important source of a highly concentrated complex of nutrients. It contains nutritional and biologically active compounds needed for the early nutrition of a newborn calf. Colostrum is produced by cows within the first 72 h after parturition, later changing to immature milk. Due to its unique composition, bovine colostrum and colostrum-based products are recently used as dietary, nutraceutical, and medicinal-supplements for the prevention and healing of a number of human diseases, for example, cardiovascular and immunity-related diseases and type-2 diabetes. Even though the variation in the content of many nutritional and bioactive compounds in bovine colostrum has been widely studied, the information on fatty acid (FA) profile changes during the first days of lactation is lacking.
The goal of our work was to optimize the extraction and derivatization of FAs from bovine colostrum and track the changes in FA profiles in colostrum and immature milk during the first four days of lactation to evaluate their possible impact on human health. Because of the complexity of the sample, comprehensive two-dimensional gas chromatography with flame ionization detection (GC×GC-FID) was used. This method provides a better separation, identification, and semi-quantification for such a complex matrix[1]. The colostrum was collected from eight Czech Fleckvieh cows on a private dairy farm (L. Klíčová, Božice, CZ), during morning milking for the first four days of lactation.
The results showed that the levels of FAs were changing over time. We have demonstrated that colostrum from later phases of lactation had a more favorable composition of milk fat from a human nutrition point of view. As each fatty acid has a different metabolic function in metabolism, the day of lactation should be considered in the production of colostrum-based products with specific compositions for nutraceutical and medicinal purposes.

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New sampling technique for determination and identification of microplastics in agricultural compartments and on its products with chemical imaging by FTIR- and Raman microscopy

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Microplastics in our environment are contaminants increasingly caused by our throwaway society. They can come into agricultural products through different processes, like the deposition of airborne microplastics, plowing under foils into the soil, or microplastics from aquatic systems used for watering. In most of these cases, microplastics accumulate on the surface of agricultural products.

A newly developed exfoliation technique with well-defined glass beads (µpeel) separates microplastics and other particles from the plant surface into a glass bead phase. This solid phase afterward is transferred into a recently developed microplastic separator for sediments (and glass beads), using flotation with air bubbles to separate even microplastics with heavy density from the aqueous solution or suspension. It enriches it quantitatively on a metal sieve plate (*µsep*) ready for further analysis [1,2].

With an FTIR microscope and Raman microscope, analytical imaging of the filter surface is processed using a recently in our group developed robust, fast, and open-source database microplastics identification software programmed with Python (µident) [3,4]. In addition, a further open-source Python-based tool (µmap) controls the image scanning of the FTIR-microscope, automatically recognizing the spot to be analyzed intensively and thus accelerating the capture of an image of the microplastics distribution on the surface of the sieve [5].

The deposition of microplastics in different in different agricultural compartments and on the surface of its products could be determined, thus revealing the possibilities of this new integral method to determine microplastics on plants.

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Food Oral Presentations

Ultra-rapid, reliable, non-targeted authentication of monofloral honey by dielectric barrier discharge ionization high resolution mass spectrometry (DBDI-HRMS).

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European beekeeping can count on a heritage of honeys that is unique in the world. Unfortunately, monofloral honey is prone to falsification and adulteration for economic profit, making the authenticity of the botanical origin, stated on the label, a long-standing concern. The botanical origin of the honey is usually certified by melissopalynological analysis combined to sensory assessments. The technique is laborious and and needs highly qualified operators. To protect the consumers and beekeepers from the risks associated with the placing on the market of products susceptible to falsification, it becomes essential to set up methods that allow a rapid, accurate, reliable, cost-effective certification. The goal of this study is the development and validation of a dielectric barrier discharge ionization high resolution mass spectrometry (DBDI-HRMS) method for the authentication of the botanical origin of monofloral honey. We analyzed a total of 112 certified samples from two independent batches. The two sample sets included honey from 7 different botanical origins (acacia, citrus, chestnut, sunflower, dandelion, linden and rhododendron). The first batch of samples included 64 samples. The independent batch was comprised of 48 samples. The samples were harvested in 2022 and originated from a variety of Italian regions. The Italian National Observatory previously verified the botanical origin of each sample by sensory analysis. After warming up at 40°C for 20 min in a thermostat, the honey were subsequently analyzed by DBDI-HRMS using a SICRIT® source (Plasmion GmbH) coupled to an Exactive Orbitrap (Thermo Fisher Scientific) by placing the open container of the honey right in front of the source for 6 seconds. The samples were analyzed in duplicate. The repeatability of the two spectral repetitions of each sample were checked by cosine similarity demonstrating their good repeatability. The spectral data were statistically analyzed and three different classifiers were built up. The random forest classifier achieved the best performances with an accuracy of 100% and 82% in cross-validation and validation with independent batch and inexperienced operator, respectively. The random forest classifier misclassified 7 samples out of 48. All the misclassified samples were characterized by unusual sensory properties due to pollen extraneous to the monofloral origin stated on the label. This study shows encouraging results that could open a new avenue for the rapid and accurate authentication of monofloral honey. For this reason, future challenges of the method will be carried out to evaluate its possible adoption in routine analysis.

Food Authentication: Mass Spectrometric Strategies for Detecting Food Fraud

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Food fraud is a multimillion-dollar business that has increased significantly in recent years and requires objective analytical methods. In this context, non-targeted metabolomics strategies using mass spectrometric techniques offer great potential. Using different examples of determining the origin of food and distinguishing between various varieties or species, it is made clear which questions can be dealt with using metabolomics techniques. The examples presented in detail include the geographical proof of origin of asparagus (*Asparagus officinalis*) and the adulteration of oregano (*Origanum vulgare*/*O. onites*) with marjoram (*O. majorana*). Starting with method development and sampling, the workflow of metabolomics studies is presented. Moreover, the identification of marker metabolites using ion mobility techniques is explained[1,2].

In addition to the application of high-resolution mass spectrometers for research and development work, the use of metabolomic strategies for routine applications is also described. The transfer of mass spectrometry-based metabolomics strategies into routine analysis is a particular challenge due to the low reproducibility of the instruments. However, it provides the basis for the methods being applicable on a large scale. Possible solutions in this context are the normalization of the data sets using so-called housekeeping metabolites, which are not subject to exogenous changes, or the implementation of a downstream process from non-targeted to robust targeted methods[1-3].

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Stable Isotope Ratio Analysis for the Authentication of Organic Wheat and Evaluation of Mycotoxin Contents and Residue Levels: From Milling to Pasta and Bakery Products

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As consumers become increasingly concerned about climate change, social inequities, and animal welfare, demand for fair-trade and organic products is on the rise. Additionally, the increasing number of cases of food fraud necessitate research for reliable analytical authentication techniques. In this work, we applied bulk and compound-specific stable isotope ratio analysis on organic and conventional wheat samples, as well as on samples of pasta, biscuits and crackers prepared using the wheat previously analysed. This way we obtained information not only about the differences in the isotope content between organic and conventional samples, but also assessed the processing effects. The organic samples in this study were collected directly from organic farms to ensure their authenticity.

For bulk analysis of C, N, S we utilized EA-IRMS, while a multi-step sample preparation process was followed for the GC-C-IRMS analysis of δ^{15}N and δ^{13}C of 9 wheat amino acids, starting by defatting the samples with a mixture of petroleum ether/ethyl ether, followed by protein hydrolysis with HCl, and amino acid purification using an ion-exchange chromatography resin. N-acetyl isopropyl derivatization was the final phase, which required acidified isopropanol for esterification and a mixture of acetic anhydride/trimethylamine/acetone for acetylation. Our findings suggest that the δ^{15}N of several amino acids could be used as organic authenticity markers, while it was noted that the processes of milling and baking had some effect on the isotope values.

Complementary to the IRMS analysis, we investigated the differences between the mycotoxins present and the residue levels in these organic and conventional samples. Trichothecenes HT2 and T2, Deoxynivalenol (DON), Zearalenone (ZEN), Aflatoxin B1 (AFB1) and Ochratoxin (OTA) were extracted via QuEChERS and were analysed by UHPLC-HRMS. Most conventional samples were found to contain HT2, T2 and DON, while the organic were less contaminated. Processing was found to reduce mycotoxin levels. Lastly, the presence of 400 residues was evaluated in the products by LC- and GC-MS/MS. Sample preparation involved QuEChERS/dSPE, and a quick polar pesticide (QuPPe) process for glyphosate.

In this multifaceted study, we highlighted the potential of compound-specific IRMS methods in organic cereal and cereal-products authentication, simultaneously providing insights into the differences in their residue and mycotoxin content.

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Spatial and temporal biodiversity of grape varieties from the Portuguese Bairrada Appellation: in the path of varietal patterns

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Sustainable viticulture and winemaking continue to represent huge challenges, where a better knowledge about the functional role of biodiversity in the vineyard and wine ecosystems is required. Particular attention should be devoted to the spatial and temporal interactions between authorized or recommended varieties for a specific demarcated region and clime and vineyard conditions (such as soil type, orientation of the lines, age of the vine, density of planting, harvesting practices, among others). Taking advantages of chemometric tools, this research aims to provide advances to examine interactions between climatic conditions, vineyard ecosystem and physicochemical data from a set of varieties cultivated in Bairrada Appellation (Portugal). Five varieties (Arinto, Cercial, Bical, Maria Gomes, and Baga *Vitis vinifera* L.) were selected as case study. For each variety, grapes from two different ecosystems were collected during two consecutive harvests. For each variety and vineyard, physical-chemical data from grapes (titratable acidity, pH and sugar content, used to estimate the technological maturity state, and free and glycosidically-potential aroma compounds were combined with edaphoclimatic data. Relationship between these three data sets and wine quality were studied using Component and Specific Weight Analysis (ComDim). Models for the prediction of grape potential for particular types of wines were developed using PLS regression. In summary, the results unveiled the high biodiversity of the Bairrada Appellation varieties, as each authorized variety presents a specific pattern, which can be expressed differently in the ecosystems under study, and through harvests. In addition, the approach used allowed to hierarchize the weight of the different variables and to estimate the adaptability of the five varieties, however, to obtain more robust data this study must be continued in time.

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PT2020 Partnership Agreement and Compete 2020 and to the Projecto Estratégico de Apoio à Fileira do Vinho na Região Centro (CENTRO-04-3928-FEDER-000001).

Fate of PFAS from soil to crop: understanding uptake and how emissions from a PFAS factory in The Netherlands impact levels in fruits and vegetables

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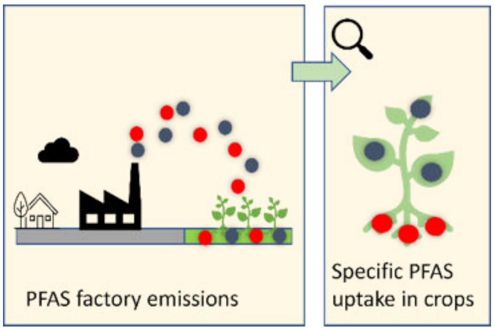
Per- and polyfluoroalkyl substances (PFAS) are a group of widely used chemical substances that persist in the environment. There is great attention to pollution of the nearby environment close to a Dutch PFAS factory, leading to increased exposures of local citizens. The consumption of fruits and vegetables (F&V) could well be an important route of exposure. Moreover, the influx of PFAS in soils and, more general, crop production systems may increase in the future through the use of reclaimed wastewater and waste materials as fertilisers. Unfortunately, little is known about the uptake and behaviour of PFAS in the soil-plant nexus.



In this study, we report on the fate of PFAS in F&V in two dimensions:

- The contamination of F&V in allotment gardens close to a Dutch PFAS factory;
- The specific uptake of individual PFAS in a root and leafy vegetable as studied in a greenhouse pot experiment.

An ultra-sensitive SPE and HPLC-MS/MS method was developed for the low and sub-pg/g detection of approx. 20 PFAS in F&V. This method was fully validated and accredited. Hundreds of F&V samples were collected near the factory by Arcadis.

The results of in total 740 samples showed significantly increased levels of PFOA and GenX in F&V from allotments close to the factory (<1 km), with concentrations up to 5 ng/g. In general, leafy crops, tubers, root-crops and cabbages had higher concentrations of PFOA and GenX compared to other F&V. Produce from allotments more distant (>1km) showed significantly lower PFAS concentrations. Given these observations, a study was conducted on the uptake of specific PFAS in more controlled conditions in greenhouse pot experiments. Radish and a leafy vegetable were exposed to PFOS, PFOA, PFHxS and PFNA through spiked biosolid-amended soils (100 ng/g). PFAS in radish roots ranged from 2.2-5.9 ng/g wet weight in the order: PFOA > PFHxS > PFNA > PFOS. A similar transfer pattern was observed for the leafy vegetable shoots of which concentrations ranged from 0.5-13.4 ng/g wet weight. PFAS accumulation varied between roots and shoots of the same plant species.

These findings show that when the production systems are contaminated with PFAS,
F&V can become important vectors for human exposure to PFAS .

Combined action of ion mobility (IM-MS) and high-resolution mass spectrometry (HRMS) to characterize known and unknown per- and polyfluoroalkylated substances (PFAS) in foodstuffs

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Per- and polyfluoroalkyl substances (PFAS) are toxic synthetic chemicals comprising several thousand compounds, which have been used since 1950 in a wide range of industrial and commercial applications (*e.g.* polymers, surfactants). Human exposure to PFAS is mainly dietary (EFSA, 2020), and infants (< 3 y.o) represent a sensitive population group. Maximum levels for four PFAS have been set for food in 2022 (European Commission, 2022). However, it remains essential to analyze and identify with certainty a wide range of PFAS likely to present a risk to human health. In this context, the Safe Food for Infants project (SAFFI, EU H2020) aims to investigate innovative technologies to enhance the detection and identification of chemical hazards such as PFAS along European and Chinese infant food chains. This work focused on both selective targeted analysis of PFAS by ion mobility mass spectrometry (Waters Synapt G2-SI, LC-IM-MS) and non-targeted screening by high-resolution mass spectrometry (Thermo Q-Exactive, HRMS) as well as data processing for specific PFAS in infant food. Twenty-eight PFAS were targeted and samples for both analyses were prepared using solvent extraction followed by SPE clean-up. IM-MS provides a collision cross-section (CCS) descriptor related to the shape and size of a molecule. $^{TW}CCS_{N2}$ values were found to be reproducible from batch to batch as expected from inter-laboratory measurements (threshold ± 2%) and a clear $^{TW}CCS_{N2}$ trend was observed for PFAS belonging to the same chemical family. A gain in sensitivity was observed using the drift time as a filter, with up to a 5-fold increase in the signal to ratio (S/N). This additional dimension combined with the mass-to-charge ratio (*m/z*) and retention time (RT) enabled the identification of targeted PFAS in food matrices with confidence, preventing false negatives due to co-eluting molecules. HRMS data were processed using Compound Discoverer SP2 (Thermo Fisher). Investigation of suspect features was made possible after estimation of the carbon number using isotopic peaks, calculation of mass to carbon (m/C) and mass defect to carbon (md/C) ratios and database searches. Data Independent Acquisition (DIA) was further used to match MS^2 profiles to suspect features. The results of the proof of concept of this method on both infant food and fish samples as well as the potential suspect PFASs highlighted will be presented.

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A strategy to evaluate food safety in circular food production systems

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The European food production system has become very effective, but resource depletion, loss of biodiversity and climate change put the system under pressure. Transitioning towards a circular production system with better waste management is one of the means to obtain a more sustainable food production system. This transition includes the re-use of nutrient containing co-products like manure, sewage sludge, crop residues, and animal by-products. However, it is known that these co-products contain chemical and biological hazards which can accumulate and recirculate in the food production system when closing production loops (van Asselt, et al. 2022 & Focker, et al. 2022). Therefore, it is relevant to identify and mitigate these hazards prior to reintroduction of the co-products in food production. In some cases precautionary measures have to be taken to prevent food safety risks.

To assess food safety in circular food systems, a framework with five questions was developed (van Leeuwen, et al. submitted). These questions include: (1) input: which co-products are used and where do they come from? (2) hazard: which hazards could be present and are they regulated? (3) fate: what is the fate of hazards in the circular food production system? (4) risk: what is the estimated potential risk by re-introducing the co-products? (5) risk management: are these risks permissible and/or can risks be mitigated? We applied this five question framework to two relevant cases: a) the application of swill for animal feed and b) agricultural reuse of treated domestic wastewater. From these cases, it became clear that there are two main knowledge gaps: first, data on the occurrence and fate of hazards and, second, the absence of fate and risk models.

For that reason a tiered strategy was developed to assess the fate of chemical hazards (Berendsen, et al. 2021). These fate studies consider the persistence and the mobility of the hazards in soil. Moreover, this strategy considers degradation products or metabolites of the parent compound and their activity, especially for non-persistent compounds. With this strategy substances are divided into three categories: (I) persistent, (II) degradable to inactive products or (III) degradable to active products. A procedure using high resolution mass spectrometry is suggested to identify the potentially formed active degradation products. We successfully applied this strategy to ten antimicrobially active substances originating from the tetracyclines, sulfonamides, diaminopyrimidines, fluoroquinolones, macrolides and lincosamides groups.

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Occurrence of high-volume production chemicals in highly consumed seafood species and evaluation of dietary intake and risk characterization

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The occurrence of thirty-five compounds in ten different species of seafood highly consumed in Catalonia, is evaluated every three months over a whole year. The compounds determined are included in the list of high-volume production chemicals (HVPC) complied by The Organization for Economic Cooperation and Development. This study focused on five HVPC groups: synthetic musk fragrances, organophosphate esters, benzothiazoles, benzosulfonamides and phthalates. A multi-residue method has been developed for their determination in seafood, using QuEChERS salt extraction and gas chromatography-tandem mass spectrometry. The results showed the presence of a large number of contaminants in the fish samples analyzed. Since consumption of seafood by the population generates a pathway of exposure to these contaminants [1, 2]. An intake exposure analysis and risk characterization study has been carried out for each contaminant. The risk characterization of carcinogenic and non-carcinogenic compounds was determined, with results of low risk.

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Polychlorinated alkanes (PCAs): Investigating levels in food from the Dutch market

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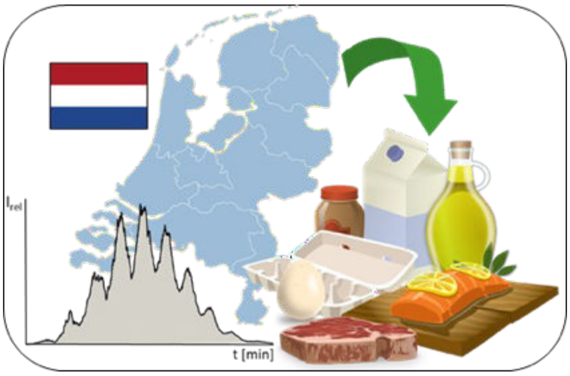
Chlorinated paraffins (CPs) are high production volume chemicals of growing concern because of their ubiquitous presence in food samples, their persistence, and their bioaccumulation potential. Their simple and cheap production established CP products as popular secondary plasticizers in PVC products, despite partial restrictions by the United Nations Stockholm Convention.



CPs are often present as very complex mixtures of primarily polychlorinated alkanes (PCAs). This complexity has led to various analytical challenges, as only an incomplete separation is possible. Additionally, availability of suitable standards has only recently begun to improve. Consequently, not much is known about the occurrence of PCAs in food or the environment. Previous market basket studies in Europe could show that PCAs are present especially in vegetable fats and oils, but also other commodities of animal and plant origin [1,2]. Thus, our goal was to set up and validate a method for analysis of PCAs in the main food groups of interest and analyse samples from the Dutch market in two stages:

(1) A survey of 55 vegetable oil and fat samples. To also account for non-mainstream products, ethnic and wholesale supermarkets were specifically included in the sampling campaign.

(2) A survey of 100 samples from other food groups of interest, including meat, fish, eggs, and milk produced in the Netherlands.

In half of the vegetable oil and fat samples, elevated PCA levels above 100 ng/g product were observed. While not all food types were included in the published studies, comparable samples are in agreement with literature data. Excluding palm oils, the mean dietary exposure through food from the Dutch market was in agreement with the other European studies. Further investigation into sources in the food production chain and also levels in palm oil refinement side streams such a palm fatty acid distillates (used for animal feed production) seems prudent.

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Food analysis: towards greener and whiter analytical methods for the determination of inorganic contaminants

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Green analytical metrics (GAC) are useful tools increasingly used to evaluate the environmental impact of analytical procedures employed in industry and research. By using these metrics, environmental impacts of analytical procedures can be identified and take actions to reduce negative effects thus contributing to more sustainable production and environmental protection. The most used green metrics generally do not fully cover what is involved in a method development that must also consider the analytical technique, validation and once in use the quality assurance of these assays. In this sense, the approach of White Analytical Chemistry (WAC) emerges as an interesting proposal with a holistic vision [1]. In this metric, 12 WAC principles were proposed as an alternative to the 12 existing GAC principles introducing several criteria that affect the quality and performance of a method from an analytical (red) and practical (blue) point of view. The RGB color model, according to which the mixture of these colors would give an impression of "whiteness" brings coherence and synergy of analytical, ecological, and practical attributes of a methodology. Being sample preparation a critical stage in the evaluation of how green an analytical method for mineral analysis in food is, an interesting metric to explore is the analytical greenness metric for sample preparation (AGREEprep) [2]. This GAC metric is based on 10 criteria that cover different aspects that contribute to the environmental sustainability of sample preparation. Official methods for analysis of the Association of Official Analytical Chemists (AOAC), that are still widely used, require concentrated acids and high temperature for matrix digestion, which is not in agreement with the principles of Green Chemistry. Drastic treatment of the matrix is not necessarily required and extraction under mild conditions may be adequate.

Several examples of alternative analytical methods for inorganic contaminants of food were developed in our research group and will be discussed [3-5] using both metrics. The proposed methods were fast, simple, and accurate for the determination of several trace elements in vegetables, milk powder and rice thus demonstrating, through this approach, that the use of drastic conditions for mineralization are not necessary to achieve reliable results for trace element determination in food.

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Pyrrolizidine alkaloids: Plant defence metabolites of serious health concern. An update on recent developments.

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Abstract

Pyrrolizidine alkaloids (PAs) are one of the largest groups of plant secondary metabolites found in nature. An estimated 6000 plant species (2% of all flowering plants) are capable of producing these compounds and over 600 different structures have been described. The main function of PAs is to protect the plant against herbivores. In recent years it has become evident that PA residues are often present as contaminants in our food and in feed, and they are also found in environmental compartments such as soil and water. Higher concentrations can result in toxic effects, causing liver damage and affecting livestock and in (rare) cases humans. Of particular concern is the fact that many PAs can act as genotoxic carcinogens, implicating that upon long-term exposure even low levels can result in adverse health effects (EFSA, 2011; 2017). Notwithstanding their widespread presence, PAs have long been largely neglected as contaminants of concern. However, in the past 15 years interest has exploded resulting in many important findings on their occurrence, their toxicological potential and in European legislation that defines maximum levels in a variety of food products.

In this presentation an overview will be given of recent developments in the analysis of PAs in connection to their presence in our food and in the environment, progress made in understanding their genotoxic potency by use of in-vitro assays and the effect of legislation and official control in reducing consumer exposure to these unwanted contaminants.

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Human dietary exposure to food packaging contaminants through *in vitro* bioaccessibility studies

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Human dietary exposure to chemical contaminants is one of the priority aspects in the food safety field in the EU and constitutes a key step in risk assessment. The evaluation of the chemical substances that can migrate from the material into the food is required to assess the safety of food packaging materials. Among the potential migrants, the Endocrine disrupting compounds (EDCs) remain a group of chemicals with implications in the occurrence of metabolic diseases with a high prevalence [1, 2].

After oral intake, foodstuffs undergo a series of processes before absorption into the body, such as gastrointestinal digestion. These processes might change the concentrations of substances available to be absorbed and could even lead to the formation of new compounds with different toxicological implications. Thus, it is mandatory to study the bioaccessibility of migrant compounds and their potential gastrointestinal degradation to understand the chemical composition of the bioaccessible fraction available for absorption.

In the framework of the BACFood4Expo project, this presentation intends to address the problem of chemical contamination of food from packaging materials and propose the in vitro bioaccessibility approach as a tool for human dietary-exposome assessment. For that, preliminary studies on the chemical behavior under different gastrointestinal digestion conditions and the bioaccessibility of food contaminants from packaging materials will be discussed [3].

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Identification of non-intentionally added substances migrating from inner coatings of metallic cans – Toward provisional risk assessment of oligoesters

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Since the decline in the use of bisphenol A, the chemistry of varnishes and coatings applied to the inner surfaces of metallic food contact materials (FCMs) is poorly documented. We hypothesised that can coatings are now diversified and bring forth various non-intentionally added substances (NIAS) to be described. The study of complex components such as NIAS requires demanding non-targeted approaches.

In a preliminary study[1], the coatings of 12 vegetable cans from the French market, where BPA is banned from all FCMs since 2015, have been investigated. Using LC-HRMS, more than 125 substances were pinpointed, among them additives such as epoxidised soybean oil and various NIAS. Mostly, 84 oligoester combinations from 8 diols and 4 diacids represented the dominant family. The stepwise organic synthesis of native and deuterated combinations of neopentyl glycol and isophthalic acid enabled a higher confidence level and monitoring in vegetable extracts. Migration of oligoesters averaged 330 µg/kg in the drained vegetables (43 to 1600 µg/kg). This preliminary study highlighted the need to carry out a proper risk assessment of this NIAS family (risk characterisation).

Further, a collaborative research project (acronym OLIGO) funded by the French National Research Agency is on-going to inform a provisional risk assessment related to oligoesters migrating from polyester-based food can coatings. One year after the project was launched, a representative set of 7 individual reference standards has been synthesised, laying the foundation for exposure and toxicological investigations. In particular, on-going work involves (i) exposure assessment by sampling ~300 canned food products with planned identification of oligoesters and semi-quantification in foodstuffs, and (ii) *in vitro* assessment of individual substances, focusing on genotoxicity, endocrine disrupting mode of action, human liver metabolism (using tritium-labelled compounds) and gastrointestinal fate.

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Non-target screening of chemicals migrating from reusable sports plastic bottles into drinking water

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Reusable plastic water bottles are popular, especially during sport, due to their lightweight and impact resistance properties. In this study, we investigated the chemical migration into drinking water stored for 24 hours in new bottles, used bottles and bottles washed in the dishwasher (Tisler and Christensen 2022). Non-target screening (NTS) by liquid-chromatography – high-resolution mass spectrometry (LC-HRMS) was performed to identify these compounds. By NTS, >400 chemicals were detected as migrating out of the plastic from new and used sports plastic bottles. After a dishwasher cycle of the same bottles, >3500 additional, dishwasher soap related compounds migrated into drinking water stored for 24 hours. In Figure 1, the compounds detected in the new bottles before and after the dishwasher cycle are shown in a heatmap. The washing cycle was not only removing migrating compounds: some plastic related compounds were migrating with higher peak intensity after the dishwasher cycle and some compounds of the dishwasher soap sticked to the material, and could not be flushed away by an additional cleaning step. The chemical structure was identified for around 50 prioritized compounds. Most plastic related compounds were plasticizers, antioxidants and photoinitiators. The highest peaks were detected for oligomers suspected to originate from the biodegradable polyester polycaprolactone. The most concerning compounds were the identified photoinitators because of potential endocrine disruptive effects. However, toxicity data were limited for most of the identified compounds in this study.

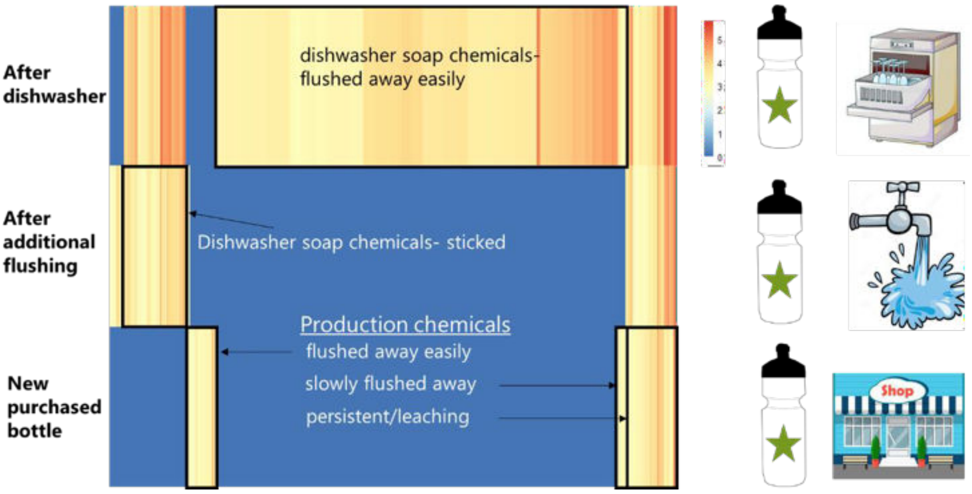

Figure 1: Heatmap of 2000 NTS compounds in the new purchased bottles. Each vertical line represents one compound; blue= not detected, the more red the color, the higher the peak intensity of the respective compound.

Reference: Tisler, S., Christensen, J.H., 2022. Non-target screening for the identification of migrating compounds from reusable plastic bottles into drinking water. J Hazard Mater 429. https://doi.org/10.1016/j.jhazmat.2022.12833

UPLC-IMS-QTOF and SPME-GC-MS to quantify migrant species in vacuum cooking bags.

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Sous Vide cooking is a fashionable technique in today's gastronomy, widely used by top chefs in some of the world's best restaurants. However, this technique involves the use of plastic intended to come into direct contact with the food under vacuum conditions, so the risk of possible hazardous compounds that may migrate from the packaging to the food should be studied[1,2].

The study presents an in-depth identification and quantification of volatile species by Solid Phase Micro Extraction coupled to Gas Chromatography Mass Spectrometry (SPME-GC-MS) and of non-volatile species by Ultra-Performative Liquid Chromatography coupled to Ion-Mobility Quadrupole Time-Of-Flight Mass Spectrometry (UPLC-IMS-QTOF) on two vacuum cooking bag materials: high density polyethylene/polyamide (HDPE/PA) and polypropylene/polyamide (PP/PA). In the case of HDPE bags, three surface configurations were studied: mixed, smooth, and embossed.

Migration tests were performed according to EU 10/2011[3] using three food simulants (acetic acid 3%, ethanol 10% and isooctane) under vacuum sealing and water immersion conditions of Sous Vide cooking. The results showed the presence of Intentionally Added Substances (IAS) such as dimethyl phthalate and dibutyl phthalate and Non Intentionally Added Substances (NIAS) such as 7,9-di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione and 3,5-di-tert-butyl-4-hydroxyphenylpropionic acid.

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Strategy for Ensuring the Safety of Novel Packaging Materials

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The safety of novel packaging materials is of utmost importance to ensure the protection of consumers as some of their constituents may be transferred to the food and result in consumer exposure. In this abstract, a generic strategy combining analytical chemistry, computational toxicology and bioassays, is presented to characterize, and possibly eliminate potential hazards with a focus mainly on the analytical strategy. The characterization starts with an untargeted compositional study aiming at (i) identifying substances present (intentional and/or non intentional) and (ii) estimating their concentration in the material, enabling a worst-case migration calculation into the foodstuffs. The methodology was based on using a headspace-GC-MS and a hexane/acetone extraction followed by screening with the complementary analytical platforms GC-MS/FID and LC-HRMS/CAD to screen from the volatile to non- volatile substances. For quantification purposes, external or internal standards were employed using universal detectors like FID and charged aerosol detector (CAD) whereas the MS was employed for the identification. After data acquisition, data processing consisted by using first a targeted approach to ensure the absence of a defined list of substances of concern. Depending on the novel material type, specific analyses are performed such as for example PFAS, screening of mineral oils, pesticides, mycotoxins or even heavy metals. If no major concern was highlighted, investigative work was carried out to identify as many substances as possible using (HR)MS data and various libraries and databases (commercial and internal) with occasionally the use of chemical standards for confirmation. And sometimes, in case of complex samples, molecular network approach was used to go beyond. Final step was the data interpretation considering regulations and for the "new" compounds (e.g. not regulated and/or NIAS), mutagenicity was excluded using toxicological predictions re-enforced by biological AMES test done in parallel and the threshold of toxicological concern approach was applied. Although it was case dependent, if a concern or hazard was highlighted several actions were taken such as performing migration tests to refine the results, to contact supplier to understand where the substance of concern is coming from to try to eliminate it or to refine the risk assessment with the support of toxicological experts. A case study of a fiber based packaging material will be presented to illustrate the strategy.

Evaluation of new approaches to decontaminate recycled polyolefins destined to come into contact with food products

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Plastic food packaging is nowadays widely used due to its many properties. However, as their use increases, there are growing concerns and worries related to both the environmental problem generated by their waste and the possible transfer of harmful substances from the plastic to the food. As a result, the search for new recycling methodologies to extend the life cycle of plastic packaging and reduce waste, as well as to guarantee the safety of recycled materials for use in food contact, is becoming increasingly important. Currently, the use of recycled polyolefins for the manufacture of these containers is not widespread, due to the fact that they have a greater diffusion capacity.

This work has focused on the evaluation of the safety of one type of polyolefin, recycled high-density polyethylene (rHDPE), and the search for strategies to achieve its decontamination in order to estimate whether its implementation in the manufacture of food packaging would be feasible. To this end, the analysis of the volatile and semi-volatile compounds present in the samples was carried out using gas chromatography coupled to mass spectrometry (GC-MS) as the analytical technique. The strategy to achieve the decontamination of flakes and pellets of this material has been based on the application of high temperatures and vacuum, obtaining a decrease in the amount of volatile compounds present in the samples, as well as in the migration experiments performed in different food simulants. It was observed that migration was analogous for 10% ethanol and 3% acetic acid, with concentrations in the order of µg/Kg. However, for 95% ethanol, a much higher number of compounds migrated and in many cases at concentrations of the order of mg/Kg

Combined untargeted screenings of substances migrating from emerging food contact materials

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The growing recognition among the general public regarding the environmental issues associated with fossil-based plastics, coupled with the rising demand for sustainability, has triggered the development of alternatives in the Food Contact Materials (FCM) sector. Bio-based and/or bio-degradable polymers, fibrous material (e.g. bagasse) and wood analogues (bamboo, palm leaf, coconut, reed) are examples of substitute materials, and they must comply with Regulation (EC) No 1935/2004. However, specific harmonized legislation is only available for plastic FCM, and the understanding of migrating chemicals remains unknown or unidentified. The study aims to bridge this gap, outlining a typical workflow that can be applied to identifying unknown compounds in emerging FCM so that a better overview of (non)-intentionally added substances can be used for safety and exposure assessments.

In the study, 99 substitute FCM were selected. Next, migration experiments were performed according to the Kitchenware guidelines[1] of the European Reference Laboratory (EURL), using simulants B (3% acetic acid) and D2 (95% ethanol) as worst-case. Afterwards, the solutions were analysed by full scan Gas Chromatography – Mass Spectrometry (GC-MS) and Liquid Chromatography – High-Resolution Mass Spectrometry (LC-HRMS). The tentative identification was performed using a workflow including spectral deconvolution followed by spectral matching with commercial mass spectral libraries. A data reduction strategy based on procedural blank subtraction was also applied to improve further data analysis.

Finally, the applicability of the developed combined approach for assessing (non)-intentionally added substances in substitute FCMs was demonstrated.

Acknowledgement

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Environment Oral Presentations

Estimation of antibiotics usage and resistance genes prevalence using Wastewater-Based Epidemiology as surveillance tool

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Antibiotic usage and ensuing antimicrobial resistance (AMR) is one of the greatest threats to public health worldwide. However, the assessment of antimicrobial agents usage in a wider community can be challenging. Wastewater-based epidemiology (WBE) is a complementary approach to current surveillance programs that overcomes some of the existing limitations such as sampling biases, long realization time, and high costs, and may help understanding the development and spread of AMR. The aim of the present study was to explore the potential of WBE for monitoring antibiotics and antibiotic resistance genes (ARGs) in influent wastewater (IWW) samples collected from the city of Castellon, Spain. Moreover, the additional analysis of antibiotics in effluent wastewater (EWW) provided information on the actual performance of the wastewater treatment plants (WWTPs) and the potential impact of the treated water on the aquatic environment. Eighteen antibiotics, most commonly prescribed, and 7 specific ARGs have been selected for their determination by liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) and qPCR, respectively. All selected ARGs, along with a total of eleven antibiotics, were identified in IWW. Highest population normalized daily loads were measured for the macrolide azithromycin followed by the quinolones ciprofloxacin and levofloxacin. While no correlations were found between antibiotics and their corresponding ARGs, certain correlations ($p < 0.05$) were identified among non-corresponding ARGs. Furthermore, data showed that only 4 of the eleven compounds detected were removed by
> 50% by the WWTP, with the exception of sulfamethoxazole *i.e.,* average removal efficiency > 80%. An additional screening by high resolution MS allowed to identify 4 metabolites WW samples. Finally, the risk that treated water could pose to the aquatic environment was also assessed, where 6 compounds indicated a potential environmental risk, exceeding the established ecotoxicological and antibiotic resistance thresholds.The results of this work provide a baseline asset for future and ongoing long-term surveillance and highlight the need of similar programs at a regional level worldwide.

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Wastewater-based epidemiology: a comprehensive public health surveillance tool

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Wastewater-based epidemiology (WBE) has become an important tool for providing evidence based assessment of public health[1][2]. WBE works on the principle that human biomarkers (e.g. pharmaceuticals and metabolites) are excreted by individuals into urban sewer networks through urine/faeces and collected at wastewater treatment plant (WWTP). As such, through the quantification of target compounds in influent wastewater (IWW) samples, it is possible to assess pharmaceutical consumption rates, health status and the lifestyle of a community for a given catchment area with high temporal resolution[3]. This is not achievable using traditional methods (e.g. surveys, sales data, prescriptions, etc.) as they contain many sources of uncertainty (e.g. bias from self-report, different report formats, incomplete treatment plan) [4]. The objective of this study was to showcase the application of WBE workflow as comprehensive surveillance tool to monitor a community in Southwest of England (1m people in 4 towns and cities). Two 24h-composite IWW samples/week were collected for one year (Mar 21–Mar 22), processed using solid-phase-extraction and analysed via UPLC-MSMS. Data analysis was conducted to assess multiple aspects of public health: (a) daily mass loads (DLs) were calculated combining concentrations and flow rates; direct disposal was highlighted using the ratio parent drug/metabolite; (b) per-capita consumption rates (population normalised daily loads, PNDLs) were obtained after DLs normalization with population served per catchment; (c) population normalised daily intakes (PNDIs) were estimated while applying human metabolism factors to PNDLs and comparing with prescription datasets [5]. Results showed that WBE can achieve near-real time monitoring, allowing the identification of direct disposal events (e.g., acetaminophen, illicit drugs), assessment of consumption rates for public health (e.g., antidepressants, cardiovascular drugs), comparison of PNDIs with prescriptions to provide a more detailed picture about public health (e.g., codeine, naproxen) and increased understanding the impact of pandemics on wider community health.

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Evaluation of antibiotic levels in a real water reuse system for agricultural irrigation

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The reuse of reclaimed water for agricultural irrigation is growing because of the increasing water stress that arid and semi-arid regions in Spain and other parts in Europe are suffering. Water reuse can alleviate the scarcity of water resources and is a continuous water source not depending on climate conditions. Regulation (EU) 2020/741 regarding the minimum quality requirements for water reuse, aims to promote the implementation of this sustainable practice in the EU. However, there is still a lack of knowledge about its potential impact on human health and the environment, associated with the presence of chemical/microbiological pollutants. Especially noteworthy is the lack of field studies, carried out under real conditions: the factors influencing contaminant transfer to the system (water-soil-plant nexus) are not well-described because studies at the pilot-scale do not always correlate with field conditions. Among the contaminants that can occur in reclaimed water, the presence of antibiotics (ATBs) is a cause for concern because they can be involved in the spread of antibiotic resistance. In this work, the occurrence of ATB was monitored in a real water regeneration and reuse system applying chlorination and used in greenhouses producing tomato. Water samples were taken in: (i) before and after (RW) the chlorination treatment; (ii) secondary reservoirs at the farms; (iii) drip irrigation points next to the plants. Soil and tomato fruits samples were taken during the growing cycle in 4 farms and were extracted using adapted QuEChERS method versions. Sample analyses were carried out by LC-MS/MS. The total concentration of ATBs in the RW was in the range 695-3735 ng/L, showing an average removal percentage of 54% after the treatment. The ATBs detected at higher concentrations were ciprofloxacin, levofloxacin+ofloxacin, azithromycin and sulfamethoxazole. In general, the concentrations found in the secondary reservoirs were comparable or lower to those observed in the RW. In the case of the droppers, ATB concentrations were similar to those of the secondary reservoirs. In soil, 10 ATBs were detected, and azithromycin, ciprofloxacin and levofloxacin+ofloxacin showed the highest concentrations (629-26998 ng/kg). In tomato, azithromycin, sulfamethoxazole and trimethoprim were detected (≤LOQ-264 ng/kg). These three antibiotics were also detected in the RW.

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Investigation of pesticides and pharmaceuticals in small water bodies using polar organic chemical integrative samplers (POCIS) and non-target screening

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Text: Pharmaceuticals and personal care products (PPCPs) are a class of organic pollutants that pose a growing concern in the context of circular economy strategies due to their potential adverse impacts on both biota and human health. This study focuses on the occurrence of PPCPs and pesticides in three small water bodies located in Norway. To assess the presence of these contaminants over an extended period, polar organic chemical integrative samplers (POCIS) were employed as sampling devices.

Over the course of one year, water samples were collected at three-week intervals and subsequently analyzed using non-target screening on Orbitrap LC-MS. Confirmation of identified compounds was achieved through the utilization of reference standards. The aim of this study was to comprehensively investigate the distribution and concentration levels of PPCPs and pesticides in the selected water bodies, shedding light on their potential impact on the surrounding ecosystem and human health.

The results of this study will contribute to a better understanding of the prevalence and persistence of PPCPs and pesticides in aquatic environments. Additionally, the utilization of POCIS in combination with advanced analytical techniques offers a valuable approach for comprehensive and time-resolved monitoring of these contaminants. The findings underscore the importance of implementing effective strategies to mitigate the release and accumulation of PPCPs and pesticides in water bodies, thereby promoting sustainable practices within the circular economy framework.

Keywords: Pharmaceuticals and personal care products (PPCPs), pesticides, polar organic chemical integrative samplers (POCIS), non-target screening, Orbitrap LC-MS, water bodies, circular economy, environmental monitoring, Norway

Tracking organic pollutants and identifying sources in urban groundwaters

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Groundwater is a source of freshwater and plays a crucial role in addressing global water demand in terms of drinking, agricultural and industrial purposes. However, in recent years, due to the growing exploitation of groundwater (GW) bodies, quality recognition and studies on the presence of contaminants in GW need to be done. A myriad of contaminants such as pharmaceuticals, personal care products, as well as artificial sweeteners, and industrial chemicals may end up in urban GW, a process which deserves further assessment [1]. The Directive 2006/118/EC amendment on groundwater quality and protection was published in October 2022 [2]. Although it includes persistent organic substances such as carbamazepine, sulfamethoxazole and 24 PFAS other persistent compounds are not included yet [2]. In our study, urban groundwater from Barcelona was analyzed using an evaporative enrichment methodology combined with LC-Orbitrap-MS in search of organic compounds used in various applications, including pharmaceuticals, industrial chemicals, and tire wear substances. As a rule of thumb, as the polarity increases (log D decreases) compounds are present in GW to a larger extent and also at higher concentrations. Mobility, among other properties, can determine the capability of a certain substance to reach GW compartments. It also has to be considered the time a compound is present without suffering changes, such as degradation or transformation, also called persistency. Similar to polarity, persistency plays a huge role in a compound being present in GW bodies due to the large reminiscence periods of this kind of water. The combination of both properties makes a compound more likely to remain in GW. In this study, the presence and occurrence of these substances and their distribution across several urban waters such as sewer, runoff and river waters have been demonstrated. Additionally, a principal component analysis (PCA) was performed with organic compound concentrations to differentiate sample groups as well as compound contributions to each cluster.

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Sample preparation investigation to extract plastic additives in agricultural soil using UHPLC-MS/MS

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Plastic additives (PAs) are chemicals blended with polymers during their production to improve the physicochemical properties of the final product. In the case of agricultural plastics (APs), certain plastic polymer types are widely used, e.g., polyolefins, containing antioxidants and light stabilizers to protect polymers against sunlight and/or ambient UV-radiation. Importantly, PAs are not covalently bound on the polymer matrix indicating their potential to leach into the environment. Considering that certain APs are directly applied into soil, namely, mulching films, and their waste mishandling, for example mulching film burying instead of collecting after use, PAs leaching into agricultural soil is highly probable, especially during AP degradation into micro- and nanoplastics (MNPs) [1]. Thus, it is of utmost importance to monitor PAs derived by APs to study their fate in soil. In this study, we investigated different sample preparation strategies to extract 19 PAs from the complex soil matrix followed by ultra-high- performance liquid chromatography tandem mass spectrometry (UHPLC-MS/MS). During the preliminary stage, the potential of plastic apparatus commonly used in the lab, for example centrifugal tubes or pipette tips, to contaminate the samples was evaluated and necessary steps were taken to avoid any background contamination. Following up, ultrasound assisted extraction (UAE) and a binary extraction combining QuEChERS and UAE principles were tested in terms of extractant, extract clean-up (based on dispersive solid phase extraction) and preconcentration to increase method detectability. To monitor the effectiveness of the two strategies, trueness, repeatability, detectability and matrix effects were evaluated. Based on the acquired performance characteristics, the application of three different workflows was necessary to effectively measure the analytes, considering their diverse polarities ($-3< \log P< 19$). In detail, aqueous UAE was proven the most efficient for polar analytes, such as dipentaerythriol, whilst UAE using hexane as the extractant achieved the best analytical performance for the analyte majority. Employing the binary extraction with ethyl acetate as the extractant was necessary to isolate Irganox 1024 and Irganox 1010, compounds that was not possible to detect without the phase separation and salting out induced by QuEChERS. All in all, effective PA extraction was achieved and validation experiments are underway to attain the completed performance characteristics for all methods.

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Unravelling the early exposure to PFAS in children at ages 4 and 8 from INMA Gipuzkoa cohort.

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The ubiquitous presence of per- and polyfluoroalkyl substances (PFAS) represents a latent threat to human health and for the environment [1]. In fact, PFAS are frequently found in human serum samples being associated with various physical alterations and behavioral difficulties in children and teenagers [2]. However, correlating health problems with PFAS exposure is often inconsistent, challenging the assessment of chronic exposure [3].

In this work, we aimed to evaluate the bioaccumulation of PFAS through the analysis of plasma samples (n=450) in children of various ages (4 and 8) coming from INMA cohort located in Gipuzkoa (Basque Country, Spain). Moreover, the links of a chronic exposure to these compounds by the drinking water supply will be assessed by analyzing current occurrence of PFAS in those cohort areas. To this purpose, a targeted High Performance Liquid Chromatography coupled to High Resolution Mass Spectrometer (HPLC-HRMS) was developed and validated to determine as many PFAS as possible in both plasma and water samples

Figure 1. Detected average values of PFAS in plasma from children at ages 4 and 8.

In addition to this targeted approach, a tiered Kendrick mass defect filter has been implemented to upgrade the identification of PFAS of different families.

The results confirmed the presence of PFAS from different families (Figure 1): FOSAs, FOSAAs, FTAs, FTUAs, PFSAs, PFCAs in plasma. Moreover, emerging PFAS such as emerging ADONA, GenX were also detected showing an early exposure to these novel compounds. The preliminary statistical analysis of the results show that were no significant differences according to sex in the exposure profiles of PFAS. The frequency of the percentage of the most detected PFAS varied slightly according to age decreasing in some cases and increasing in others (Table 1).

Table 1. Frequencies of detection above the method limit of quantification (MLOQ) of the most detected PFAS.

COMPOUND	% > MLOQ	
	4 YEARS	8 YEARS
PFOA	98.6	86.7
PFOS	98.6	98.8
PFHxS	81.6	84.2
PFNA	94.2	91.3
PFDA	76.3	72.1
PFUdA	67.6	37.9

Regarding drinking water analysis, the compounds were found at pg/mL levels including PFBS, PFHxA, PFHpA, ADONA, PFOA, FHpPA, PFNA, PFDoA, PFUdS, PFDoS, FOSA, PFTrDS, N-MeFOSE. The sum of the 20 PFAS included in the European Water Directive 2020/2184 did not show results above the specified limit (0.1 μg/L) in the areas studied suggesting that drinking water might not be the principal source of exposure in this case.

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Trace element speciation made easy: introducing Frontal Chromatography ICP-MS

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The speciation analysis of trace elements has attracted much attention as their total concentration fails in describing and predicting their behavior in and impact on environmental and biological systems [1,2]. Present speciation methods [3–5] are commonly based on the hyphenation of separation techniques like HPLC (High Performance Liquid Chromatography), GC (Gas Chromatography) or IC (Ion Chromatography) to sensitive detection methods such as Atomic Fluorescence and Absorption Spectrometry or Inductively Coupled Plasma – Mass Spectrometry (ICP-MS) [3,4,6,7]. This approach requires two separate instrumentations (chromatography plus high sensitivity detector, with associated purchase and running costs) and generally long analysis time: what is most important is that they exceed the needed resolution when a two-species problem is tackled, as often true in speciation analysis (e.g., Cr(VI) *vs.* Cr(III), As(V) *vs.* As(III), inorganic *vs.* organic Hg). Such shortcomings have been recently addressed in our research group by introducing the innovative Frontal Chromatography ICP-MS approach (FC-ICP-MS)[8]. Species separation is achieved by positioning a short (2-6 cm), home prepared, low pressure (50-75 μm particle diameter) column packed with the adequate stationary phase between the peristaltic pump and the nebulizer in the traditional, commercially available ICP-MS configuration, avoiding the use of an HPLC system. The advantages of this approach (easiness of operation, associated fast extraction, reduced analysis time), its validation in biological and environmental samples (fish tissues, natural waters and human hair) along with the successful protocols setup up to date (determination of As(III) and As(V) [8], Cr(VI) [10] and methylmercury [RIF], plus antimony redox speciation [RIF]) will be presented and discussed. Limitations and possible future development will be also presented.

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Thermal processes and secondary recycling regulate the atmospheric levels of highly toxic polychlorinated naphthalenes in an urban Mediterranean site

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Although production of industrial- and pesticide-grade Persistent Organic Pollutants (POPs) has been prohibited since early 00's, residues are still present in all environmental compartments and illicit usage is still documented at a global scale[1]. Island of Cyprus comprises a unique location for exploring air quality over the broader Eastern Mediterranean and Middle East (EMME) region[2]. The present work extends previous reports[3-5] on the occurrence and fate of a large panel of highly toxic compounds in the atmosphere over Cyprus and explores comprehensively the atmospheric distribution of polychlorinated naphthalenes (PCNs) and polybrominated diphenyl ethers (PBDEs), both listed under the Stockholm Convention text[6]. Total (gas/particulate phase) concentrations of PCNs were measured similar to urban locations in the broader region[7]. Contrarily, the corresponding levels of PBDEs were similar to remote locations in Greece and Turkey[8, 9]. Regressions of logarithms of partial pressure against ambient temperature for PCNs revealed that secondary recycling from contaminated terrestrial surfaces regulates their atmospheric variability. Enthalpies of surface-air exchange (ΔH_{sa}) for PCNs were significantly correlated to vaporization enthalpies (ΔH_v) determined by chromatography, corroborating presence of short-range revolatilization sources. Homologue concentration ratios of PCNs and ternary diagrams suggested inputs from thermal processes all over the sampling period (Jan-May 2018), whereas potential influence of Aroclor technical mixtures cannot be excluded. An inverse pattern for PBDEs was observed. The corresponding regression slopes were shallow, implying long-range atmospheric transport, whereas ΔH_{sa} were insignificantly correlated and close to ΔH_v, suggesting that, unlike PCNs, volatilization sources for PBDEs are of minor importance. The Dachs-Eisenreich equilibrium model predicted well the gas/particle partitioning quotients (K_p) of PCNs, whereas the Li-Ma-Yang steady state model predicted better the partitioning behavior of PBDEs. The temperature-dependent quantitative structure-property relationship (QSPR) model, calibrated by our research team, exhibited equal performance in predicting K_p and can be used as reference in future studies.

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Passive Sampling Of High Production Volume Chemicals And Polycyclic Aromatic Hidrocarbons In Outdoor Air Samples. Application And Risk Assessment Evaluation

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The impact of air quality on human health is a growing concern, with a rising number of individuals being affected by respiratory infections, lung cancer, and heart disease [1]. As a result, it is crucial to monitor and control the presence of air pollutants.

Air samples are typically collected using active sampling methods, such as high volume samplers. However, an alternative option is passive sampling, which offers advantages such as cost-effectiveness, noise-free operation, minimal space requirements, and no need for a power supply [2]. Nevertheless, in order to ensure the reliability of the passive sampling, it is crucial to optimize sampling time and determine the diffusive uptake rates for each specific target compound.

In this study a passive sampling has been applied and evaluated for a group of contaminants incluiding polycyclic aromatic hydrocarbon (PAHs) and high production volume chemicals (HPVCs), such as benzothiazoles (BTHs), benzenesulfonamides (BSAs), phthalate esters (PAEs), organophosphate esters (OPEs), ultraviolet stabilizers (Tinuvins) and phenolic antioxidants (PAs). Pressurized liquid extraction (PLE) was used as extraction technique of the analytes from the passive sampler and gas chromatography-mass spectrometry (GC-MS) to determination them.

Monitoring study was carried out at six urban siter close to the two petrochemical parks of Tarragona (Spain). The compounds with the highest concentrations were PAEs and PAHs, with arithmetic means of of 27.2 ng·m^{-3} and 22.9 ng·m^{-3}, respectively. Non-carcinogenic and carcinogenic risk assessment have been estimated, and PAHs were the target compounds with the highest contribution.

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Development of a novel passive sampler based on membrane assisted solvent extraction and molecularly imprinted polymer for monitoring of selected pharmaceuticals in surface waters

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This work demonstrates development, evaluation and pre-liminary application of a novel passive sampler for monitoring of selected pharmaceuticals in environmental waters. The samplers were calibrated in laboratory-based experiments with the view to obtain sampling rates (Rs) for carbamazepine, methocarbamol, etilefrine, venlafaxine and nevirapine. Passive sampling was based on the diffusion of the target pharmaceuticals from surface water through a polypropylene membrane bag which housed an ionic liquid as a green receiving solvent and a molecularly imprinted polymer. Effects of biofouling, deployment time and receiving solvent type were optimized for selective uptake of analytes in surface water. Notably, there was a decrease in the uptake of selected pharmaceuticals and consequently a decrease in their sampling rates in the presence of biofouling. The optimum matrix-matched obtained sampling rates ranged from 0.0007 - 0.0018 L d^{-1} whilst the method detection and quantification limits ranged from 2.45 - 3.26 ng L^{-1} and 8.06 - 10.81 ng L^{-1}, respectively. Deployment of the optimized passive sampler was done in a dam situated in the heart of a typical highly populated township in the Gauteng Province of South Africa. Out of the five targeted pharmaceuticals, only etilefrine and methocarbamol were detected and quantified at maximum time weighted average concentrations of 12.88 and 72.29 ng L^{-1}, respectively.

Investigating the sorption capacity of four Nigerian clays for the removal of arsenic, cadmium, chromium, and lead from water

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Contamination of water bodies with potentially toxic elements (PTE) arising, for example, from mining sites and industries has made surface water unsafe for human consumption in many parts of the world. Remediation of such polluted water is a major challenge, especially for rural communities in Africa. Due to the high cost of sophisticated water treatment techniques, there is considerable interest in the use of sustainable locally-sourced alternatives. Clay has been reported to be a good adsorbent for remediation of PTE-contaminated water due to its high cation exchange capacity, high pore volumes, and large surface area (1). Clays are cheap, readily available and non-toxic to handle (2).

In this study, the sorption capacities of four different clays, Eme, Ihievbe, Imeh and Inereh obtained from Owan West, Owan East, and Etsako East Local Government Areas in Edo state, Nigeria were individually investigated. Batch sorption experiments were performed and As, Cd, Cr and Pb concentrations determined using an ICP-MS instrument (Model 7700x, Agilent Technologies, UK). The effects of contact time, pH, analyte ion concentrations, and adsorbent dosage were considered with a view to establishing optimum sorption conditions. The four clays all demonstrated high adsorption capacity for Cd, Cr and Pb with over 90 % removal from solution. In contrast, As removal was < 50 %. Equilibrium was reached within 20 minutes. Sorption was influenced by pH, analyte concentration and adsorbent dosage.

Further studies will be carried out to establish the adsorption isotherms and kinetics of the sorption processes. The clay will be characterised with XRD, SEM and FTIR to determine their elemental composition, morphology, and structure.

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Determination of haloacetic acids in reclaimed water and drinking water by direct injection and hydrophilic interaction chromatography coupled with mass spectrometry

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Chlorination is one of the most widely used tertiary treatments. Nevertheless, chlorine together with dissolved organic matter generates the so-called disinfection by-products (DBPs). Haloacetic acids (HAAs) are included within the DBP category and due to their known carcinogenic and mutagenic effects, the EU Drinking Water Directive [1], US-EPA [2] and WHO guidelines [3] regulate the levels for some of these compounds in drinking water. The sum of dichloroacetic acid (DCAA), trichloroacetic acid (TCAA), dibromoacetic acid (DBAA), monochloroacetic acid (MCAA) and monobromoacetic acid (MBAA) are commonly known as HAA_5, and should not exceed 60 µg/L in drinking water. In addition, EPA's Contaminant Candidate List (CCL) [4] includes bromochloroacetic acid (BCAA), bromodichloroacetic acid (BDCAA), dibromochloroacetic acid (DBCAA), and tribromoacetic acid (TBAA) as known or suspected contaminants that are expected to occur in public water systems and may require regulation. This work presents a fast and simple method based on the direct injection of samples to determine nine HAAs in reclaimed wastewater (WW) and drinking water (DW) treated by chlorination, as an alternative to traditional and slow solid-phase extraction (SPE) methods. The determination was carried out by liquid chromatography coupled to triple quadrupole tandem mass spectrometry (LC-QqQ-MS/MS) using the HILIC mode. To our knowledge, this is the first methodology that analyzes the aforementioned 9 HAA by DI using HILIC. The proposed method was successfully validated in the two mentioned matrices at two concentration levels (10 and 100 µg/L). Mean recoveries were in all cases in the range 84- 113% (*n*=4) with relative standard deviations (RSD) ≤21% (intra-day and inter-day precision, *n*=3), with limits of quantification (LOQ) between 0.25 and 25 µg/L. The applicability of the method was demonstrated by analyzing: four chlorinated WW, fifteen DW, and one chlorinated groundwater sample. Concentrations detected were in the range of 0.4-9.0 µg/L for drinking water and 89-2491 µg/L for chlorinated WW samples.

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Chemometrics for improved identification and quantification of polymers in blood using Pyrolysis-GC-MS

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A recent study has found the presence of micro and nano-plastic particles (MNPs) in human blood, raising questions about the extent of human exposure [1]. To study this exposure, analytical methods need to be optimized to maximize sensitivity and selectivity due to the low concentration of MNPs in blood. In this study, chemometrics tools were applied to pyrolysis-gas chromatography mass spectrometry (Py-GC-MS) data to improve the identification and quantification of MNPs in human whole blood. The current quantification method uses a single polymer marker compound [1]. However, pyrolysis can produce a complex mixture of products that are not always unique to a specific polymer. Additionally, compounds can be susceptible to co-pyrolysis, further complicating the quantification process by making it dependent on the specific polymers present in the sample.

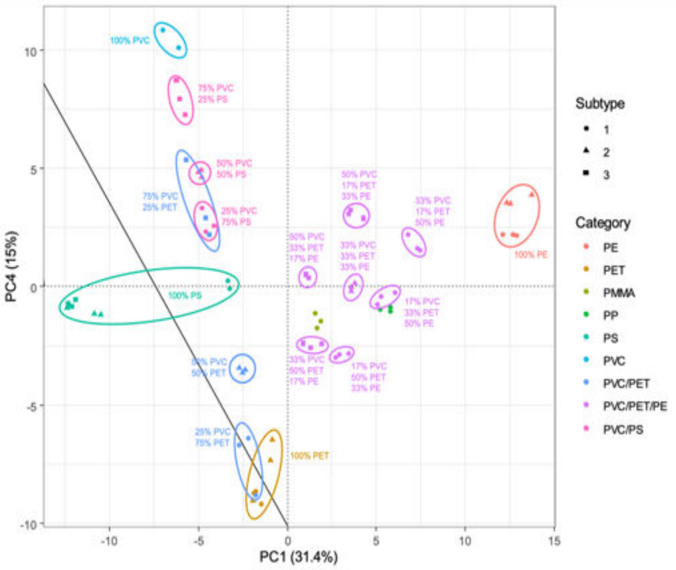

Figure 1: Classification and quantitation of polymer mixtures using PCA

A validated Py-GC-MS method was used to analyse references and spiked matrix samples. PARADISe software, which is a PARAFAC2 based deconvolution and identification tool was used to resolve untargeted Py-GC-MS data [2]. Preliminary exploratory data analysis using PCA (Figure 1) shows that variables selected using PARAFAC-2 are capable of discriminating polymer types and mixtures, Supervised methods (e.g., SVM, random forest and regression trees) are currently being optimised to develop more accurate classification and calibration models.

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Analysis of Micro- And Nanoplastic in Human Blood

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Recent research has found micro and nanoplastic (MNP) particles in blood, this discovery has raised concerns about human exposure to internal microplastics [1]. Although the effects of these particles on human health are not well understood, MNPs have been detected in drinking water, food, and air, suggesting widespread exposure. As plastic usage continues to increase, the levels of microplastics in our environment are expected to rise in the future [2]. To better understand and quantify MNPs in biological samples and study their effects and interactions, robust analytical methods with high sensitivity and selectivity are needed.

In this study we present a robust and sensitive sample preparation and analytical method using pyrolysis-gas chromatography coupled with mass spectrometry (Py-GC-MS) and apply it to measure plastic particles in human whole blood from healthy volunteers. Blood was subjected to enzymatic digestion using proteinase K [3], followed by sequential microfiltration on 0.7 µm and 0.3 µm glass filters. To ensure the accuracy of the method, it was validated using a quality control (QC) sample spiked with six polymers: polyvinyl chloride (PVC), poly(methyl methylacrylate) (PMMA), polypropylene (PP), polystyrene (PS), polyethylene (PE), and polyethylene terephthalate (PET).

The method was validated through spiking experiments, and the pyrolysis parameters and GC separation conditions were optimized to improve specificity and sensitivity. The results showed that PMMA, PE, PET, and PVC were identified, and PE, PET, and PVC were quantified in the blood samples. The method proved effective in extracting, identifying, and quantifying low concentrations of major polymer types found in plastics. To ensure reproducibility and accurate recovery of polymer particles in Py-GC-MS analyses, it is imperative to implement quality control measures.

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Towards simultaneous size determination and polymer identification of nanoplastics

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With tremendous amounts of plastic waste being disposed to the environment, micro- and nanoplastic (MNP) pollution is a major environmental concern. Therefore, the need for comprehensive and reliable analytical methods for both identification and quantification of MNPs is indisputable. However, whereas for larger microplastic particles this problem is relatively easier to solve, with numerous well-established protocols already, the submicron nanoplastic particles remain largely unperceivable due to sextremely low mass concentrations and technical difficulties posed by their minute size.

A common approach in nanoplastic analysis is using mass-based analytical approaches, such as pyrolysis-gas chromatography with mass spectrometry detection (Py-GC/MS). This technique allows identification of polymers present in the sample. However, Py-GC/MS cannot distinguish between sizes. Therefore, any result obtained may potentially stem from either a low number of large particles or a huge number of minute particles, which is a serious limitation while investigating overall fate of plastic litter in the environment.

One technique allowing for size separation is asymmetrical flow field-flow fractionation (AF4). While separating particles below 1 µm based on their sizes, it does not hive hint about their chemical composition. Including both AF4 (as the first dimension) and Py-GC/MS (as the second dimension) in one workflow seems thus to be an extremely promising approach.

In this light, we investigate the potential for indirect coupling AF4 with Py-GC/MS on effluents from several Dutch wastewater treatment plants as exemplary samples. Large amounts (thousands of litres) of effluents are pretreated by crossflow ultrafiltration. Typically described AF4 methods for nanoplastic fractionation were adjusted to be compatible with the pyrolysis step and mass spectrometry. Since during fractionation sample is highly diluted, we preconcentrate our samples in-line with a modified injection, thus lowering the LOD by a factor of 1000. We further examine influence of in-between freeze-drying of the fractionated samples, such as compatibility with instrumentation, manipulating detection limits and analysis time. Our results suggest that using both analytical techniques together results in versatile and complementary information on the tiniest plastic particles present in the environment.

So far, no direct AF4-Py-GC/MS coupling has been described. We strongly believe that our research largely contributes to achieving this in the future, providing the community with an advanced and comprehensive tool for nanoplastic analysis, and thus – to properly evaluate the emerging nanoplastic pollution problem.

Towards FAIR data in the field of environmental research on microplastics

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Scientific research operates on the basis that the results are disseminated through scientific publications. These publications generally do not contain all the original data or do not contain them in a format that is reusable. In some fields, such as medicine, the limitations of this system of operation have been openly acknowledged (e.g., in the words of the European Medicine Agency's former executive director, "relying solely on the publications of clinical trials in scientific journals as the basis of healthcare decisions is not a good idea. Drug regulators have been aware of this limitation for a long time and routinely obtain and assess the full documentation (rather than just publications)" (doi:10.1016/j.ejim.2019.11.022pmid: 31948774)). Does this mean that scientists and regulators in other fields are left with publications that are inherently of limited value? The need to change the situation lead, as early as 2016, to the provision of guidelines to improve the Findability, Accessibility, Interoperability, and Reuse of scientific data (doi:10.1038/sdata.2016.18). Since then, many initiatives have emerged and some funding agencies even promote the publication and sharing of data.. Examples include the European Union's programme for science funding already requiring almost all data to be FAIR and he USA government announcement that research articles and underlying data generated with federal funds should be publicly available at no cost by the end of 2025 (doi:10.1038/d41586-022-02820-7). In practice, however, what looks simple and obvious might rapidly become difficult to implement. Since open science, when properly implemented, is synonymous with "pragmatic solutions to understanding, disseminating, scrutinising, and implementing research findings" (doi:10.1136/bmj.p1609), we propose here an example of a ready-to-use approach in the case of microplastics' in environmental compartments. Publications on microplastics have exploded in recent years, a significant part of them dealing with their detection and measurement in air, soil and water. It has been repeatedly pointed out that this research suffers greatly from the lack of agreement on the standardisation of sampling and analytical methods used. Less appreciated is the difficulty in accessing the original data and the disparate way in which it is published. Less than a third of the articles in the field published since 2016 contain a data sharing statement and data were explicitly shared via a data repository in only 13.8% of the articles (doi:10.3389/fenvs.2022.912107). Clearly, the first goal to achieve is that all data are accessible but equally important is that data are reusable (i.e., accompanied by the corresponding metadata). This communication is a step forward in this direction.

Microplastics in urban freshwaters: a case study in the city of Amsterdam

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Microplastic (MP) contamination of freshwaters has been globally established [1]. Ranging from macro to nano-sizes, plastics enter aquatic ecosystems via direct sources such as discharges from wastewater treatment plants and drainage outlets, and indirect sources such as atmospheric deposition, surface runoff, and human activities [2]. In the environment, plastics undergo degradation and weathering processes depending on their physicochemical properties and environmental conditions. The aim of this study is to determine the abundance of MPs in the urban region of Amsterdam, the Netherlands, which includes a large network of canals and waterways. Two sampling campaigns were conducted during the summer of 2022 and winter of 2023 at 11 locations in the Amsterdam canal network using a state-of-the-art in-situ volume-reduced sampling pump. The qualification and quantification of the polymers for the size fraction of 25-300 µm were performed in two laboratories at VUA (Amsterdam) and ENPC (Paris) using two complementary analysis methods, pyrolysis-gas chromatography-mass spectrometry (Py-GC-MS) and micro-Fourier transform infrared (µ-FTIR) imaging. The canals in the city center exhibited the highest concentrations of MPs, reaching up to 1,673 MP particles/m^3 and 782 µg/m^3. PP, PE, PS, and PET were the most abundant polymer types found. The findings revealed comparable trends in MP abundances using both analysis techniques while also providing complementary information regarding concentrations based on mass and number of particles.

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Poster Presentations

P01 - Cholinesterase Biosensor for neurotoxin monitoring based on highly stable Prussian blue-copper mediator electrodeposited on diazonium modified surfaces

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The neurotoxins both natural and insecticides can be monitorised based on their cholinesterase global inhibition. The Prussian blue electrochemical mediator is very sensitive for the detection of hydrogen peroxide or thiocholine but its usage for biosensor development is hampered by its poor stability at neutral pH that is required by the enzymes. Several strategies have been proposed such as covering the electrode with a membrane to prevent the mediator dissolution or development of mixed Prussian blue configurations that contain also other cations such as copper. We present modified electrodes based on diazonium chemistry to produce surfaces with high affinities for Prussian blue for improved stability. The mediator stability was further improved by the usage of metal nanoparticles tested into two configurations: as seed for modified Prussian blue precipitation or on the electrode surface before electrodeposition of the Prussian blue, only the latter option ensures also an efficient transfer of electrons to the electrode. The stabilized Prussian blue allowed reproducible signals for hydrogen peroxide while the analytical signals recorded with a classical Prussian blue are slowly decreasing from one measurement to the next (fig 1).

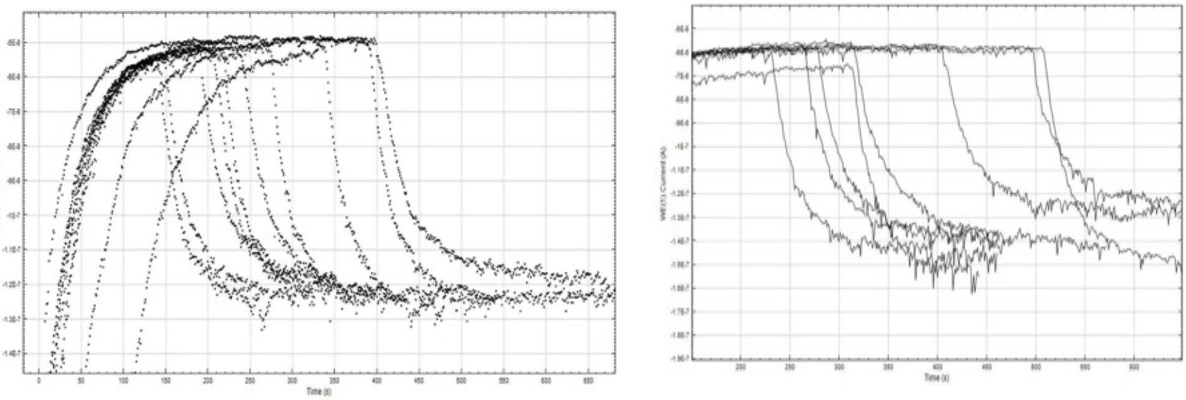

Fig. 1 Analytical signals obtained for hydrogen peroxide using stabilized (lef) and classical Prussian blue (right) electrodes.

P02 - Solid-liquid–solid-phase extraction coupling (SL–SPE) established for the analysis of levoglucosan in atmospheric particulate matter

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Detection of the extent of anthropogenic or natural atmospheric pollution with regard to ambient air quality in urban and remote locations, and therefore the risks of exposure to humans and the environment gains increasing focus in times of growing environmental protection interests. Particularly, the large amounts of wood smoke emissions from, *i. a.,* residential wood combustion or wildfire events, are major sources of particulate matter that contains toxic, mutagenic, or carcinogenic compounds, such as polycyclic aromatic hydrocarbons. Possible adverse health effects of exposure to wood smoke can affect, *i. a.*, the nasal mucous membranes and lungs. Regarding effects on the environment, global and regional climate change is also linked, *i. a.*, to black carbon and brown carbon emissions from biomass burning due to their interference with solar radiation.

For monitoring atmospheric pollution, the most relevant and frequently used organic molecular tracer in wood combustion is levoglucosan (1,6-anhydro-β-D-glucopyranose, LV). LV is a glucose-based anhydro sugar, and therefore a simple and fast extraction step with a medium polar solvent can be applied to extract this very soluble compound from airborne particulate matter (PM) collected on glass fibre filters. A newly established filter sample extraction process presented here combined direct coupling of a solid-liquid (SL) sample extraction unit with a solid-phase extraction (SPE) cartridge (SL–SPE). It was tested on PM samples with an aerodynamic diameter smaller than 10 µm. Compared to other known extraction methods for the analysis of LV in PM material from aerosols, the SL–SPE method offers advantages in saving time and resources.

The SL–SPE sample extract analysis was performed by gas chromatography–mass spectrometry on trimethylsilylated LV in combination with a stable isotope dilution assay. Testing the SL–SPE performance to selectively analyse LV in PM on glass fibre filters provided good recovery of 97%, with standard deviation lower than 5%. Verification of the SL–SPE extraction efficiency showed that the percentages of LV amounts detected by second extractions were lower than 0.07% of those from first extractions. The LV amounts detected in exemplary PM samples, collected at two urban background sites during winter season, ranged from 7 to 748 ng m^{-3} ambient air.

P03 - Identification of Persistent, Mobile and Toxic compounds in runoff water samples of Barcelona

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Abstract:

The escalating threat of climate change and water scarcity calls for the exploration of alternative water supply options like groundwater. Nevertheless, groundwater is overexploited in many cases and the use of rainwater to recharge it is contemplated. However, it is crucial to acknowledge that surface runoff water often contains dissolved contaminants. Unfortunately, these contaminants may not be effectively removed through infiltration mechanisms and can persist in receiving waters. Additionally, identifying the sources and reducing inputs of these pollutants can be very difficult and expensive [1]. In light of these challenges, the scientific community is actively working towards establishing the concentrations of these pollutants in various water bodies, as well as new methodologies [2]. To address this knowledge gap, the application of high-resolution mass spectrometry (HRMS) is imperative. By utilizing HRMS, scientists can study unknown compounds thoroughly and perform retrospective analyses to identify compounds of interest. In this study, our focus was on optimizing a method combining gas chromatography quadrupole time-of-flight (GC-QTOF) and liquid chromatography quadrupole time-of-flight (LC-QTOF). This method offers the advantage of determining a wide range of compounds and their related unknown substances present in runoff water. Given the polarity of various compounds investigated, sample preconcentration was achieved through evaporation. Overall, this research aims to contribute to a better understanding of the pollutants present in stormwater runoff and provide insights into effective strategies for their identification and mitigation.

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P04 - Multiresidue method for the determination of antibiotics and their metabolites in sewage sludge and soils

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Antibiotics are a group of pharmaceutical compounds widely used to prevent and treat bacterial infections in human and veterinary medicine. After their administration, they are excreted through urine and/or faeces as parent compounds and/or metabolites. Due to the extended use, overuse and misuse of antibiotics, parent compounds and their metabolites are continuously being discharged to the environment mainly through effluents from wastewater treatment plants. They can also be accumulated onto sewage sludge generated during wastewater treatments [1–2]. This fact is of special concern when sewage sludge is applied onto agricultural soils as fertilizer [3]. In this work, a method for the determination of twelve antibiotics and eight of their metabolites has been developed and validated for their determination in composted sludge and soil. The method involves ultrasound-assisted extraction, dispersive solid-phase extraction clean-up and analytical determination by liquid chromatography-tandem mass spectrometry. Variables affecting each stage of the analytical method were thoroughly optimised. Method quantitation limits were in the range from 0.03 to 7.5 ng g^{-1} dw. Precision, expressed as relative standard deviation, was lower than 17% for all the compounds (mean value: 8.3%). Average accuracy, expressed as relative recovery, was in the range from 87.8 to 106%. A preliminary application of the method revealed macrolides and fluoroquinolones as the antibiotics at the highest concentrations in both types of samples.

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P05 - LC-MS/MS method for the determination of chiral β-blockers, antidepressants and two of their metabolites in soil and sludge

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More than half percent of pharmaceuticals in use are chiral compounds which are commonly administered as racemates. Their enantiomeric fractions can be altered after administration by metabolism and by selective sorption and/or microbial degradation once excreted and released to the environment [1]. As enantiomers can exhibit different ecotoxicity, a proper environmental risk assessment should involve their enantiomeric determination [1,2]. Nevertheless, most of the studies about the environmental occurrence, fate and risk assessment of chiral pharmaceuticals do not consider their enantiomeric composition. This fact can be explained by the great challenge to overcome when developing analytical methods for multi-residue enantiomeric determination of pollutants because of their identical physico- chemical properties [2]. In this work, an analytical method was optimised and validated for the simultaneous extraction and enantioselective determination of chiral β-blockers (n=3), antidepressants (n=5) and two of their metabolites in agricultural soils, compost and digested sludge. Sample treatment was based on ultrasound-assisted extraction and extract clean-up by dispersive solid-phase extraction [3]. Analytical determination was carried out by liquid chromatography-tandem mass spectrometry using a Chirobiotic V chiral column [3]. Enantiomeric resolutions were in the range from 0.71 to 1.36. Accuracy was in the range from 85 to 127% and precision, expressed as relative standard deviation, was lower than 17% for all the compounds. Method quantification limits were 1.21–52.9 ng g^{-1} dry weight (dw) in soil, 0.76–35.8 ng g^{-1} dw in compost and 13.6–90.3 ng g^{-1} dw in digested sludge.

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P06 - New Passive sampling method by POCIS and Sample Preparation method by SPE for the analysis of Perfluorinated compounds (PFAS) in river and tap waters.

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Perfluorinated compounds (PFAS or PFC) are a family of molecules composed of fluorocarbon chains of variable length and a functional group such as carboxylic or sulfonic acid. They have been widely used since the 1950s in many products, such as in fire-fighting foams, for non-stick and hydrophobic coatings, or as surfactants. Their composition makes them particularly chemically resistant, and they therefore tend to accumulate in organisms and in the environment. Perfluorinated compounds have come under increased scrutiny in recent years because of their suspected adverse effects on human health. Solid Phase Extraction (SPE) based on polymers has proven to be effective for PFAS analysis as it can be used for concentration and quantitation, or as passive sampling tools such as POCIS (Polar organic Chemical Integrative Sampler).

In this work, a list of 13 PFAS were analyzed by passive sampling using AFFINIMIP® POCIS PFAS in river water. The experiment was realized over a period of 7 days and the method of analysis was LC-MSMS. Very good results were obtained in river water, as the linearity of the adsorption for the 13 compounds was demonstrated over a period of 7 days. The Sampling rates (Rs) calculated were between 0.083 and 0.223 L/day.

The cartridge format was also demonstrated for trace analysis. 500 mL of tap water was spiked with 10 PFAS at 24ng/L and then passed through a polymeric SPE cartridge to concentrate the PFAS prior to LC-MSMS analysis. For this study, the SPE procedure was both tested manually and with an automaton. During the analysis, a delay column was used to avoid contamination of PFAS coming from HPLC device and reach low limits of detection. Very good results were obtained in tap water with recovery yields ranging from 88% to 102%. The mean reproducibility of the method (relative standard deviation obtained over eight samples) was 4.3% (with a maximum of 12% for PFOS).

P07 - Analysis of persistent and mobile toxic chemicals in human urine by mixed-mode liquid chromatography

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Persistent and mobile toxic chemicals (PMTs) have drawn attention nowadays due to their particular properties in comparison to other common environmental contaminants. However, the very polar nature of PMTs makes their extraction from aqueous samples and subsequent analysis very challenging. Therefore, PMTs are overlooked in human monitorization works, limiting the knowledge of the human exposome. In that sense, the need for comprehensive analytical methods to determine PMTs in human samples has arisen [1]. In this work, several sample treatment procedures as well as analysis conditions were studied to determine almost 40 PMTs in human urine using liquid chromatography (LC) coupled to tandem mass spectrometry (MS/MS). Regarding sample preparation, 12 procedures consisting of non-discriminatory (i.e., dilute-and-shoot (DS) and freeze-drying) and discriminatory methods (i.e., mono- and multi-layer solid phase extraction (SPE) and liquid-liquid extraction (LLE) variants) were evaluated. For the analysis, 3 chromatographic modes were studied, such as reverse-phase, HILIC and mixed-mode. Under optimal conditions, DS and a combination of SPE using polymeric Oasis HLB cartridges and salt-assisted LLE were selected as optimum procedures since they allowed the detection of the maximum number of PMTs (>70%). For the analysis, mixed-mode LC provided a greater capacity for retaining the targets. Therefore, both procedures were validated at 5 ng/mL and 50 ng/mL in urine in terms of limits of quantification (< 5 ng/mL), linearity (> 0.9990), trueness (70 – 130 %) and precision (> 70 %). As a first screening approach, a pooled urine sample (n=5) was submitted to DS and analysed quantifying diverse PMTs [2]. For examples, industrial chemicals (acrylamide and bisphenol S), biocides and their metabolites (2-methyl-4-isothiazolin-3-one, dimethyl phosphate, 6-chloropyridine-3-carboxylic acid, and ammonium glufosinate) and an artificial sweetener (aspartame) were determined at ng/mL levels. The outcomes of this work showed that humans are also exposed to PMTs due to their persistence and mobility, and therefore, further human risk assessment is needed.

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P08 - Comparison of Trace Element Deposition in Cupressus macrocarpa Leaves and Soils from a High- Pollution Area in the Puchuncaví Valley (Chile)

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In central Chile, located in the Puchuncaví Valley (PV), is found one of the most important and oldest industrial complexes (ICs), containing several industries and the most important activity is the Cu smelter and refinery. This mineral plays a fundamental role in Chile's economy, the country being among the world's leading Cu producers [1]. However, several environmental problems associated with Cu mining activity are widely known, particularly with trace elements, such as Cu, Zn, Pb, Cd, and As [2-4]. This study assesses the enrichment factors of As, Cd, Cu, Pb, Sb and Zn, both in soils and in *Cupressus macrocarpa* leaves from high-pollution-load area such as the PV (Chile).

The undisturbed soil was sampling with a monthly periodicity, between April and July 2021 [3] and the leaf samples were collected from the annual growth of the trees in April 2021 [2]. Both leaves and soil were collected from five different monitoring sites located around the IC in the PV, each sampling sites represent different degree of pollution impact. The elemental analyzes both leaf and soil were analyzed according to a previous study [2,3]. The enrichment factors (EFs) in soil was evaluated according [3] and Fe was chosen as reference element. The EF in leaves were evaluated according [2]. The leaves showed EFs of: As (33-188%) Cd (0-85%), Cu (96-607%), Pb (55-341%), Sb (45-142%) and Zn (28-264%). These results can be interpreted as, EF<50% normal condicons, EF of 50–100% moderate enrichment, and EF> 100% significant enrichment. On the other hand, the soils showed EFs of: As (0,7-3,0), Cd (all samples were below limit of quancficacon, BLQ), Cu (0,7-37), Pb (0,7-5,7), Sb (all samples were BLQ) and Zn was possible to quancfy in the site closest IC (EF of 1,3). The results can be interpreted as: EF<2 minimal enrichment, 2<EF<5 moderate enrichment, 5<EF<20 significant enrichment and, 20<EF< 40 very high enrichment. The highest EF both leaves and soils correspond to sampling sites closest IC. The EF observed in soils and *C. macrocarpa* leaves collected in the Puchuncaví Valley, was clearly affected by anthropic accvices to a variable degree, depending on the distance to the IC.

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P09 - Pollution source of Trace Element in *Cupressus macrocarpa* Leaves from a High-Pollution Area in the Puchuncaví Valley (Chile)

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In central Chile, located in the Puchuncaví Valley (PV), is found one of the most important and oldest industrial complexes (ICs), containing several industries such as natural gas terminal, cement, chemical companies, coal-fired power plants, and Cu smelter and refinery [1]. This study assesses the source of 23 trace elements in *Cupressus macrocarpa* leaves from high-pollution-load area such as the PV.

The leaf samples were collected from the annual growth of the trees in April 2021 and were collected from five different monitoring sites located around the IC in the PV [2]. The trace elemental was analyzed according to a previous study [2]. The trace elemental sources were assessed through of Principal component analysis (PCA) with varimax rotation according [2]. The leaf PCA with varimax rotation showed three common factors contributing to 78.6% of the total accumulated variance. The first factor, with 34.8% of the variance, grouped elements such as Na, Mg, Al, Ti, V, Fe, La, and Ce. The association of these elements might be attributed to mineral–crustal sources. Due to the low La/Ce (0.7 to 1.1) and La/Nd (1.7 to 2.4) ratios that are consistent with impacts from the resuspension of local dust [3]. Furthermore, the association of lanthanides with other elements such as Al, Na, Ti, Fe, and V, recognized as natural in soil from the PV [4], suggests that the association of these elements in leaves may be attributed to natural sources. The second factor (25.5% of the total variance) grouped elements such as Cu, Zn, As, Mo, and Pb. The association of these elements might be attributed to anthropogenic sources represented by copper smelting, as reported in previous studies on the area [2,4]. The third factor (F3) (18.3% of the total variance) grouped elements such as Mn, Co, and Ni. These elements in Puchuncaví soil are attributed mostly to the coal-fired power plants. Elements as Ni has shown high concentrations of fly ash from coal-burning power plants [5], Co and Mn can be found in high concentrations in coal-derived sources [6]. It is probable that these elements in the leaves have anthropic sources such as the coal-fired power plants of the IC. The trace element source observed in *C. macrocarpa* leaves collected in the PV, was clearly affected by anthropic ac`vi`es.

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P10 - Identification of 1,3-diphenyl guanidine and 1,3-di-o-tolylguanidine metabolites produced by *in-vitro* human liver metabolism

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1,3-Diphenyl guanidine (DPG) and 1,3-di-o-tolylguanidine (DTG) are rubber additives widely used for the vulcanization of tires, among other uses. Due to their large-scale use, these compounds have been detected in the environment and the recent studies suggest that DPG is an endocrine disrupting chemical and both chemicals are linked to allergic dermatitis. Nevertheless, only few studies have assessed the potential human exposure to them[1], while little is known about their metabolites. Thus, in this work, Phase I and II *in-vitro* biotransformation of DPG and DTG has been investigated using human liver microsomes (HLMs) and cytosol (HLCYT). Samples were analyzed using liquid chromatography coupled to quadrupole-time-of-flight mass spectrometry (LC-QTOF). Finally, suspect screening combined with non-target screening approaches were employed to identify the potential metabolites. Four phase I and two phase II metabolic products were identified for DPG. Hydroxylation of the benzene ring led to the identification of mono- and di-hydroxylated DPG (DPG-227 and DPG-243). Subsequently, secondary phase I reactions resulted in the formation of a hydroxy- phenylurea product (DPG-228), and a hydroxy-cyclic derivative (DPG-225). Additionally, N-glucuronidation (DPG-387) and O-glucuronidation products (DPG-403) were also identified. As regards DTG *in-vitro* metabolism, the 4 analogous phase I metabolites and the 2 phase II metabolites were identified: mono- and di-hydroxylated DTG (DTG-255 and DTG-271), the hydroxy-tolylurea product (DTG-256), the hydroxy-cyclic derivative (DTG-253) and the N- and O-glucuronides (DTG-415 and DTG-431). For both compounds, different isomeric products were recognized. Finally, a suspect screening with pooled urine samples allowed the tentative identification of a DTG phase I metabolite. Further research is undergoing in order to confirm the presence of these metabolites in more samples and to determine the extent of exposure to DTG and DPG.

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P11 - Identification of Persistent Mobile and Toxic Contaminants in Environmental Water Samples using a Suspect Screening Strategy

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Persistent, mobile and toxic substances (PMTs) are becoming a matter of concern in the aquatic environment due to their intrinsic properties which led, in recent years, to an increased detection in the water cycle. This growing relevance has drawn the attention of the European authorities, placing these substances at the same level of concern as persistent, bioaccumulative and toxic (PBT) chemicals under REACH. The NEPMTUNE project (www.nepmtune.webnode.es) aims at identifying and studying the behavior of PMTs in Spanish aquatic environments, as well as to propose strategies for their mitigation. Within the project activities, a suspect screening strategy was designed to investigate the presence and sources of PMTs in different aqueous matrices (river water, raw and treated wastewater, urban run-off and landfill leachate). A suspect screening library, containing ca. 1100 chemicals, was created using a prioritization strategy based on toxicity (T), persistence (P) and mobility (M) criteria. Then, 31 samples collected in Spain in 2023, from different type and geographic origin were submitted to the entire protocol, employing lyophilization, as concentration step, and mixed-mode liquid chromatography coupled to quadrupole-time of flight mass spectrometry (MMLC-QTOF-MS). So far, the preliminary results permitted the identification of more than one hundred PMTs. The most frequently detected compounds (detection frequency >50%) were chemical reagents, such as tetrabutylammonium and 1,3-di-o-tolylguanidine and pharmaceuticals, such as alprenolol, metformin and desmethylvenlafaxine. The results of this work provide a baseline asset for future monitoring and for the development of water treatment technologies for better management and reduction of risk caused by these compounds.

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P12 - Investigation of the potential biotransformation of different pharmaceuticals in zebrafish (*Danio rerio*) embryos, utilizing LC-TIMS-HRMS combined with suspect and non-target screening workflows

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Emerging contaminants have garnered great scientific interest since pharmaceuticals and their transformation products, among other xenobiotics, are frequently detected in the aquatic environment. Further investigation is required due to potential risks to the aquatic environment and organisms [1]. Zebrafish (Danio rerio) embryo (ZFE) has emerged as a powerful model-organisms in ecotoxicological studies since it poses similar biotransformation systems to mammals. Biotransformation is crucial in toxicokinetic studies, but the identification of biotransformation products (bio-TPs) presents many challenges [2]. LC-HRMS offers a powerful approach to detecting and identifying these bio-TPs, while hydrophilic interaction liquid chromatography (HILIC) could be used for orthogonal identification of bio-TPs in addition to reversed-phase liquid chromatography (RPLC). Trapped Ion Mobility (TIMS) is a promising technology for the separation of isomers and could provide additional experimental evidence to enhance identification confidence.

This study aimed to investigate the biotransformation capacity of ZFE exposed to different pharmaceuticals and to identify the tentative bio-TPs, utilizing LC-HRMS. Furthermore, it highlights the importance of using two chromatographic techniques, and TIMS technology, to identify candidate bio-TPs. For the extraction of pharmaceuticals in ZFE samples, organic solvents were added, and a bead-beating homogenization process was followed. The ZFE extracts were analyzed by RPLC and HILIC, in both positive and negative ionization, by LC-ESI-QTOFMS. The extracts were also analyzed with LC-TIMS-QTOFMS, which introduced an additional dimension of separation. Target-screening approach was followed for the identification of the parent compounds, whereas suspect and non-target screening workflows were applied for the identification of bio-TPs. Overall, bio-TPs from both oxidative and conjugative metabolic reactions were identified. The analytical data of this study, highlight that RPLC and HILIC, in combination with collision cross-section (CCS) values, provided additional evidence to support the identification of bio-TPs.

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P13 - Identification of volatile and semivolatile compounds in primary packaging of ophthalmic formulations by GC-MS

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Primary packaging of ophthalmic formulations is usually plastic, which is more flexible than glass with high resistance to breakage. In general, the elaboration of ophthalmic formulations in the form of eye drops is carried out in multi-dose plastic containers due to their practicality and low cost. Thus, the safety of these medical materials should be evaluated.

In the present work, non-targeted analysis using gas chromatography coupled to mass spectrometry (GC-MS) was used to investigate volatile and semivolatile compounds present in different components of these medical materials. The samples were characterized by FTIR mainly as polyvinylchloride (PVC), Silicone and High-density polyethylene (HDPE). The analysis of volatile compounds was performed at 70 ºC and 30 min by Purge and Trap (P&T), and semi-volatiles after the extraction with methanol at 70 ºC for 24 h. Each analysis was made in triplicate and using two internal standards. Among volatile compounds determined by P&T, alkanes, alkenes, and aldehydes were the most abundant substances, since they can be polymerization residues and oxidation products. Cyclohexanone was identified in uni-dose container samples, which was also previously described in other saline packaged PVC for clinical use [1, 2]. In the methanolic extracts, some carboxylic acids and their ester derivatives like tetradecanoic acid, hexadecanoic acid or hexadecanoic acid, methyl ester, were identified, as well as some plasticizers like bis(2-ethylhexyl)terephthalate or erucamide, which is a slip agent used in polyolefins.

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P14 - Chemical and Effect-Based Analysis of Per- And Polyfluoroalkyl Substances in an Indirect Potable Reuse System During Aquifer Recharge

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Indirect potable reuse applications through soil aquifer recharge (SAR) offer great potential to alleviate water scarcity in light of the growing effects of climate change and the corresponding increased pressure on conventional freshwater sources. SAR enables not only to replenish groundwater levels but also produces in general high quality potable water. However, while such applications offer great advantages, there persist concerns regarding the fate and behaviour of very persistent and very mobile compounds. In that regard, especially per- and polyfluoroalkyl substances (PFASs) need to be critically evaluated as they are ubiquitously detected in the environment and are linked to a series of potential adverse environmental and human health effects. Their analysis in environmental samples however remains challenging and especially ecotoxicology studies are scarce.

This study presents the first preliminary results of an in-deep analysis of an indirect potable reuse system in the Netherlands, where treated wastewater affected surface water is used for aquifer recharge. In June 2023, surface water samples (n = 2) were collected at the start of the indirect reuse system and numerous groundwater samples (n = 14), which cover varying soil depths, from 2 to 31 m below the surface, and water traveling times from 34 to 380 days. Samples were analysed by a combination of target chemical analysis and effect-based tools. Specifically, samples were analysed by liquid chromatography–mass spectrometry for a set of 47 target PFASs and further extended by a novel *in vitro* chemical activated luciferase gene expression (CALUX) assay to study their potential mixture effects.

Therefore, the applied approach enables not only to gain a deeper understanding about the fate and behaviour of various PFASs during soil passage, but also accounts for their potential ecotoxicological effects in mixture. Overall, the results will help to gain a deeper understanding of indirect potable reuse applications and how very persistent and very mobile compounds might affect such systems.

P15 - Solving the PFAS Challenge: Comprehensive Screening of Environmental Samples against 1000s of Compounds in a Single Run

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Introduction

PFAS are known as “forever chemicals” due to their persistent, bio-accumulative, toxic (PBT) properties and ubiquitous presence in the environment and organisms. Ca. 5000 PFAS are marketed worldwide, making their systematic environmental monitoring an extremely challenging task. On top, there is a lack of reference standards or spectral libraries, and there are plenty of isomers. Adding trapped ion mobility spectrometry (TIMS) to UHPLC-HRMS allows for comprehensive monitoring of organic micropollutants in environmental matrices such as water and soil.

Objective

Complete solution for PFAS characterization in environmental samples, combining ion mobility supported target analysis with wide scope non-target screening against a database of 5000 compounds.

Methods

Water samples were spiked with sets of PFAS compounds or taken from common household and environmental water resources. Data independent acquisition was performed on a high-resolution mass spectrometer equipped with ion mobility. Kendrick mass defect analysis filtered potential PFAS from the matrix background, based on the fluorine content (repeating CF_2 units). Spectra were compared with either a spectral library of available PFAS standards in a targeted workflow or with the Norman network and NIST databases of 5000 entries for non-targeted analysis. From the Norman and NIST databases, information for the exact mass, in-silico fragmentation, and CCS prediction for every PFAS compound was derived which were used for an automated and untargeted identification of PFAS.

Results

The ion mobility feature of the system was utilized for several purposes. First, it could separate coeluting isobars and isomers. Second, the TIMS filter resulted in higher sensitivity and lower detection limits of the targeted PFAS as well as significantly higher quality of full-scan MS and bbCID MS/MS spectra. Finally, collisional cross sections (CCS) as additional identification criteria enhanced the identification confidence with was based on retention time, exact mass, diagnostic fragmentation ions and the isotope pattern fit.

Discussion

The wide-scope suspect screening of real-life samples against the NORMAN network database proved to be a comprehensive approach for a fast and efficient identification and quantification of PFAS against the total set of 5000 compounds in complex environmental matrices. Therefore, it will assist in understanding the chemical universe of PFAS in the environment and protecting environment, wildlife, and human health.-Bruker Confidential-

P16 - Application of new state-of-the-art HG-ICP-MS/MS methods for (sub)ppt speciation analysis of some less studied TCEs (Ge, Sb and Te) in natural waters

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Despite the proven toxicity of some chemical elements such as lead, cadmium or arsenic, the measurement of their concentrations in natural systems has for many years been considered solved (with the exception, for a time, of their chemical speciation) and the focus of environmental analytical chemistry has been essentially on the determination of organic pollutants. This situation has changed recently with the development of new information technologies and, above all, with the need to shift the current economic model towards decarbonisation. This combination has made much of the periodic table topical. It is worth noting in this respect that the fifth technical assessment (2023) of critical raw materials for the EU considers some 50 chemical elements to be technologically critical! The various stakeholders involved then realised that there were still many gaps in our ability to measure the concentrations of many of these elements and their species at the concentration levels needed to study and understand natural processes. We will present here the case of elements such as antimony, germanium and tellurium, which are present in natural waters in concentrations of (sub)ppb or even ppt. Their analysis by ICP-MS, the standard technique in many laboratories, is further complicated by unfavourable properties such as high ionisation potential, element isotope number distribution, isobaric and polyatomic interferences, and intricate chemistry. The development of advanced methods based on the hydride generation (HG) technique, combined with state-of-the-art ICP-MS/MS detection, offers an answer to the challenge, but requires careful sampling, preservation and laboratory work and is hampered by the absence of certified reference materials and standards of some species. The new methods will be discussed through their application to natural waters of contrasting chemistry in different seasons.

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P17 - Alleviation of β-lactam antibiotic pollution by using β-lactamase entrapped agarose discs: Reusability and scale-up studies

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Abstract

Antibiotic pollution in the environment has direct role in the rise and spread of antibiotic resistance. To evade this problem, there is a necessity of development of sustainable remediation technique to lower the level of antibiotic residues from the environment. Among various antibiotic hydrolysing enzymes, β-lactamases are note-worthy, as they are known for hydrolysing broad spectrum of β-lactam antibiotics. Considering this, in the present study, β-lactamase from *Bacillus tropicus* EMB20 was entrapped in agarose matrix and thereafter used for remediation of meropenem (a carbapenem antibiotic) and other β-lactam antibiotics. The lactamase entrapped agarose discs completely hydrolyzed 50 mgL^{-1} meropenem within 30 min of treatment. The used discs were successfully recovered and reused for up to 5 cycles with 72% antibiotic removal efficiency. The major hydrolyzed product of meropenem was found to be opened-lactam ring structure with m/z 402.16. The hydrolyzed product(s) were observed to be non-toxic as revealed through microbial MTT, confocal microscopy, and growth studies. Moreover, the discs simultaneously hydrolyzed mixture of antibiotics containing 50 mgL^{-1} each of meropenem, doripenem and amoxicillin after 30 min of treatment. The treatment process was scaled up under continuous mode using fixed-bed column bioreactor packed with layers of lactamase entrapped agarose discs and sand gravels. The mixture of 50 mgL^{-1} of each antibiotic were observed to be completely removed after 20 min of retention time in the FBCR. The setup could be reused up to 5 cycles with 41-45 % removal efficiency of antibiotic mixture. Overall, the study shows the potentiality of using this lactamase entrapped agarose discs in sustainable abatement of antibiotic pollution from the contaminated environmental samples.

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P18 - Determination of glyphosate and aminomethylphosphonic acid in human urine

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Glyphosate represents one of the most used herbicide since early 1970s, while aminomethylphosphonic acid (AMPA) is its main degradation product in the environment and certain crops. Glyphosate is commonly employed in weed control, and also used as a desiccant prior to harvest of e.g. cereals and oil seeds. People are exposed to glyphosate and AMPA through dietary intake and possibly through environmental routes (dust/air born particles). Human biomonitoring is a way to assess the aggregated internal exposure.

In the present study, the aim was to develop a sensitive and selective method for determination of both glyphosate and AMPA in human urine. The investigation consist of a derivatization step using trifluoroacetic anhydride (TFAA) as reagent, followed by gas chromatography (GC) coupled to tandem mass spectrometry (MS/MS) analysis. Negative chemical ionization (NCI) is employed as ionization interface between GC and MS/MS. The derivatization procedure allows to reduce the polarity of the analytes and increase the MS detection capability. In order to perform quantification, two isotopically labelled internal standards (ILISs), namely glyphosate-d_2 and AMPA-^{13}C-^{15}N, are added to the 50 µL human urine aliquot used for sample analysis.

The method herein reported can be considered as an alternative to methods based on liquid chromatography coupled to MS/MS. The advantages of the GC-MS/MS based method are: uses a smaller volume of urine; no cleanup required; the sensitivity is not prone to ion suppression.

The method was validated according to SANTE/11312/2021 guideline and a limit of quantification of 0.1 µg L^{-1} was achieved for both analytes.

P19 - Ultra-Sensitive High-Throughput Methylmercury Detection in Human Hair via Streamlined ICP-MS analysis

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Mercury species pose a significant threat to life forms in the biosphere, with their interactions with the environment and their impact on human health being specific to each species. Methylmercury (MeHg) exhibits higher toxicity and bioaccumulation potential compared to inorganic species (iHg), making fish consumption, particularly of top predator species, a critical pathway for MeHg exposure and a global public health concern [1]. Traditionally, MeHg exposure in fish consumers is monitored by measuring total Hg (or, more rarely, MeHg) concentration in blood, serving as a biomarker. However, recent studies have shown that MeHg accumulates in human hair at concentrations up to 250 times higher than in blood, offering a valuable and less invasive alternative biomarker for MeHg exposure assessment [2]. The MeHg levels in human hair can vary widely (ranging from tens of µg/kg to a few mg/kg) based on individual dietary habits. To meet the specific demands of this application, highly selective and sensitive analytical speciation methods are required for exposure monitoring. Here, we present a novel, simple speciation method based on ICP-MS that enables selective and rapid determination of MeHg in biological samples (fish tissue and plankton). The method employs a custom-made column filled with a strong anion exchange resin to effectively block interfering iHg species before analysis in the ICP-MS system. In contrast to an earlier protocol developed by our research group [3], this improved approach eliminates the dilution step after Hg species extraction, allowing the analysis of very low concentrated samples. An optimized HCl-HBr-thiourea mixture is used for exhaustive extraction, followed by direct analysis after a 15-minute ultrasound-assisted extraction. The developed method demonstrates excellent performance, with a detection limit of 2.8 µg kg^{-1} in solid samples (based on a 20 mg sample mass) and negligible interference from iHg, even in cases where iHg:MeHg ratios are as high as 50, which is uncommon for human hair samples. To validate the procedure, a standard reference material of human hair (NIMD-01, from the National Institute for Minamata Disease) was used, yielding accurate results. Furthermore, the proposed method was successfully applied to 49 human hair samples, revealing a strong correlation between MeHg concentration and individual weekly fish and seafood intake. These results highlight the effectiveness of this streamlined and easily implementable method for high-throughput MeHg exposure monitoring.

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P20 - Biodegradability testing approaches at Nouryon

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The impact of chemicals on the environment, climate change and the integrity of the biosphere has become increasingly important in recent decades. Because of these consequences, knowledge of the biodegradability of chemicals is crucial. Biodegradation of chemical compounds refers to the reduction in complexity of the chemical through the metabolic activity of microorganisms. Under aerobic conditions microorganisms oxidize organic substances and convert them into carbon dioxide, water, and biomass (growth–linked biodegradation).

At Nouryon we investigate the biodegradation of many chemicals and collaborate with industrial and academic partners to not only predict and study the behaviour of chemicals in the environment, but also to gain knowledge that will enable us to develop new environmentally friendly products.

We use the biodegradation guidelines currently established by the Organization for Economic Co-operation and Development (OECD, 1992). The OECD testing levels (ready and inherent biodegradation, and simulation tests) are integrated into the European legislation and play a role during chemical risk assessment and decision-making.

Here we present the main testing approaches of our biodegradation laboratory at Nouryon, and we show biodegradation results of a selected set of chemical compounds produced at our company. First, we investigated the biodegradation behaviour in a ready biodegradation test (OECD 301 D or OECD 301 F), and if complete mineralization is observed within four weeks, the test substance can be classified as readily biodegradable. Secondly, inherent biodegradability tests (OECD 302 A, 1981) are performed when negative results are observed in ready tests because inherent tests are less stringent and therefore there is a greater chance of observing biodegradation. Inherent biodegradation tests are useful to demonstrate that a chemical is not persistent in nature (ECHA 2017b).

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P21 - Determination of Persistent, Mobile, and Toxic compounds in soil and crops by mixed- mode liquid chromatography coupled to tandem mass spectrometry.

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Session: Persistent and mobile toxic chemicals (Environmental Chemistry)

The current scenario of climate change will increase the demand for water reuse for different uses, including agricultural practices. However, the reuse of, for example wastewater causes new concerns about the possible presence of emerging contaminants, particularly persistent, mobile, and toxic (PMTs) compounds. Consequently, the irrigation with such contaminated water may lead to an uptake of PMTs by soils, crops and plants. The determination of these polar compounds (Log D < 3.5) is challenging and requires advanced analytical methodologies. In the present study, mixed-mode liquid chromatography coupled to tandem mass spectrometry (MMLC-MS/MS) has been used for the determination of eight PMTs in escarole (*Cichorium endivia*) and soil. The selected PMTs (Benzophenone-3, Clarithromycin, Imazalil, Metformin, Sulpiride, Terbutryn, Tiapride and Tramadol) were separated within 10 minutes using mobile phases consisting of water, acetonitrile (ACN) and ammonium acetate (variating between 5 - 15 mM along the gradient). MS/MS, using a triple quadrupole mass analyzer, operated in positive-ion mode. The acquisition of at least two transitions per compound allowed accurate quantification and reliable identification of each PMT. Sample treatment of both soil and escarole was performed by optimizing a QuEChERS-based method. Briefly, 10 g of escarole and 5 g of soil were extracted with 10 mL ACN and the citrate kit salts ($MgSO_4$, NaCl, $Na_2HCitrate \cdot 1.5H_2O$ and $Na_3Citrate \cdot 2H_2O$). In the case of escarole, this procedure was followed by a clean-up step using $MgSO_4$, PSA and C_{18}. Finally, for both matrices, the extract was 2-fold diluted and 5 µL was injected in the MMLC-MS/MS system. Results showed satisfactory overall recoveries (including extraction and matrix effects) in a range of 70-120%. The methodology was fully validated at the Limit of Quantification (LOQ) and 10 times LOQ level considering the following parameters: linearity, precision, and accuracy. Finally, preliminary results of a large sample campaign of escarole and soil samples irrigated with spiked water will be presented.

Acknowledgements
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P22 - Towards a comprehensive 4D-targeted workflow suitable for environmental and human biomonitoring of xenobiotics leveraging LC-VIP HESI-TIMS-HRMS.

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Thousands of chemicals are dispersed into the environment and may threaten ecosystems and health. Through environmental and human biomonitoring, the chemical fingerprint of the exposure of living organisms to xenobiotics can be revealed. Today, thousands of chemicals can be reliably detected and quantified through wide-scope target screening using high- resolution mass spectrometry coupled with chromatography. However, the sample complexity results in high matrix effects and, thus, may hamper the detection of compounds at low concentration levels. Reliable detections are also prevented by the complexity of MS/MS data produced by data-independent acquisition (DIA) modes. Therefore, ion mobility spectrometry and the derived collision cross section (CCS) values can be leveraged to LC-HRMS target screening as a new dimension aiming at better separation and more reliable identification of xenobiotics in complex matrices.

Environmental and human biospecimens were spiked with semi-polar to polar xenobiotics and treated using generic sample preparation protocols, resulting in extracts suitable for LC-HRMS wide-scope target screening. The extracts were analyzed using LC-TIMS-QTOFMS by an optimized broad mass and mobility transfer bbCID mode (DIA). Analyses were implemented both with ion mobility switched on and off, aiming to unveil the benefits of trapped IMS (TIMS) dimension to LC-HRMS. Moreover, the possibility of improved ionization efficiencies was investigated through the use of vacuum-insulated probe-heated electrospray ionization (VIP-HESI) source compared to the typical ESI source. Data treatment was performed using Bruker's TASQ 2023 and TargetScreener 4D platform.

The results of this study confirmed more than two times higher sensitivity with the VIP-HESI source for half of the tested analytes. This is crucial for the detection of analytes at low concentration levels, a pain point in the trace analysis of environmental and human biospecimens. Moreover, the TIMS dimension improved signal-to-noise ratios, whereas the ion mobility filtering provided higher-quality chromatograms and full-scan MS and bbCID MS/MS spectra, since the background signal caused by co-eluting isobaric analytes and matrix components was largely deconvoluted. The criterion ΔCCS% was added to the established screening parameters of mass accuracy, retention time shift, isotopic pattern fitting and the presence of qualifier ions. Therefore, the incorporation of TIMS in the established LC- HRMS targeted workflows steer towards increased overall identification confidence due to the reduction of false positive and negative results. -Bruker Confidential-

P23 - Organic microcontaminant levels in treated and reclaimed water from wastewater treatment plants in southeastern Spain: are they ready for future EU requirements?

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In the European Union (EU), wastewater treatment is regulated by Directive 91/271 (and its modifications) [1], which is currently under revision and update, and where requirements are expected for organic microcontaminants (OMCs). The focus is put on the percentage OMC removal that might be required in the future, with the Swiss requirements as main reference (80% of selected OMC removal from the influent to the tertiary effluent) [2]. In this context, one of the aims of the EU LIFE-PHOENIX project is to evaluate the performance of 8 WWTPs, which apply tertiary treatments, to remove OMCs and how far these technologies are from a possible target (80% of removal). A total of 233 OMCs (including pharmaceuticals, pesticides, antibiotics and hormones) were monitored in secondary and tertiary effluents (reclaimed water) to establish a removal percentage during the treatments. Two samplings were carried out in summer and winter. The target OMCs were analysed by liquid chromatography coupled to triple quadrupole mass spectrometry (LC-MS/MS) using the direct injection (DI) technique. Only for hormone analysis, solid-phase extraction was used as sample pretreatment. As expected, the total concentration of OMCs occurring in the secondary effluent was reduced after the tertiary treatment. However, the different workflows applied in the WWTPs (transport, storage, etc.) and the different sampling strategies used (punctual samples in tertiary effluents and composite samples in secondary effluents), made not possible to establish unequivocal contaminant removal rates. Despite this, an average reduction of 45% could be estimated when chlorination was applied as a tertiary treatment, and somewhat higher, when it was combined with ozonation (76%). The average total concentration of OMC varied depending on the sampling period: generally lower in summer than in winter (higher consumption of pharmaceuticals, especially antibiotics).

References:

[1] Council Directive 91/271/EEC of 21 May 1991 concerning urban waste-water treatment.
[2] Water Protection Ordinance (WPO) of 28 October 1998 (Status as of 1 January 2021).

Acknowledgements:

The authors are grateful for the contribution of the LIFE PHOENIX project, funded by the European Union under Grant Agreement No. LIFE19-ENV/ES/000278.

P24 - Evaluation of the generation of trihalomethanes in reclaimed water produced by advanced chlorination tertiary treatments using headspace and gas chromatography coupled to mass spectrometry

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Among the different tertiary treatments available to produce reclaimed water (RW), chlorination is widely used as an easy and cheap treatment. However, chlorine can react with dissolved organic matter to generate disinfection by-products (DBPs) such as trihalomethanes (THMs). In this work, the combination of chlorination and an advanced oxidation processe, solar photo-Fenton, was studied to minimize THM generation. Despite THMs are the predominant DBP group generated in disinfection processes, they are only regulated in RW in Italy [1]: the sum of chloroform (CFM), bromodichloromethane (BDCM), dibromochloromethane (DBCM) and bromoform (BFM) should not exceed 30 µg/L. In this work, a method has been developed for the analysis of the four regulated THMs by headspace (HS) extraction and determination by gas chromatography coupled to mass spectrometry (GC- MS). HS extraction parameters (temperature, sample time and volume, stirring speed, HS syringe penetration and temperature) were optimized to achieve maximum sensitivity. The method was successfully validated at two levels (250 and 2500 µg/L). All mean recovery values (*n*=4) were in the range 93-119%, with precision values (expressed as relative standard deviation, RSD) ≤14% (intra-day) and ≤15% (inter-day) for both levels (*n*=3). LOQs were set at 250 ng/L and the linear range was 250-2500 ng/L ($R^2 \geq 0.9900$ in all cases). The validated method was applied to the analysis of secondary effluents from wastewater treatment plants (WWTP) treated by (a) conventional chlorination (NaOCl) and (b) chlorination combined with solar photo-Fenton. The four THMs were detected when chlorination was used, and the highest concentrations were 24.7 µg/L (DBCM) and 26.5 µg/L (BMF). When chlorination was combined with solar photo-Fenton, only CFM was detected and at extremely low concentrations (0.6 µg/L). The results confirmed that chlorination combined with solar-photo- Fenton, can reduce the formation of THMs and encouraged to improve conventional chlorination processes applied in water regeneration.

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P25 - The occurrence of pharmaceuticals in three different WWTPs in Saudi Arabia and their potential effects on groundwater

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Abstract

Despite increasing interest in pharmaceutical emissions worldwide, studies of environmental contamination with pharmaceuticals arising from wastewater discharges in Saudi Arabia are scarce. Therefore, this study investigates the occurrence of 11 pharmaceuticals from different therapeutic classes in three wastewater treatment plants (WWTPs) in Riyadh city, Saudi Arabia. One hundred forty four collected samples were extracted and analyzed using either solid phase extraction (SPE) followed by liquid chromatography-tandem mass spectrometry (LC-MS/MS) or ultrasound-assisted extractions (UAE) combination with LC-MS/MS for wastewater and soil samples, respectively. The average concentrations in the influents and effluents were generally higher than their corresponding concentrations reported in previous studies. The highest concentrations measured in the influents of these WWTPs were for caffeine and acetaminophen (> 1000 µg L^{-1}), followed by ciprofloxacin, metformin, ofloxacin, diclofenac, atenolol, cephalexin, trimethoprim and baclofen (< 1000 µg L^{-1}). Baclofen has been reported for the first time in the aquatic environment, at concentration ranging between 0.33 and 2.82 µg L^{-1} in WWTP influent.

Mass loadings in the inputs and outputs of the detected compounds in the larger WWTPs were significantly ($p \leq 0.5$) higher than for the small scale WWTP. There were very few significant correlations between the physiochemical parameters of the collected samples, the operational parameters of the WWTPs, and the observed concentrations of the studied pharmaceuticals in the studied WWTP influents and effluents.

The overall average removal efficiency was high (≥80), with no significant difference ($p > 0.05$) between the treatment technologies. Acetaminophen and caffeine were almost eliminated in all three WWTPs. The samples collected in the cold season generally had higher levels of detected compounds than those from the warm seasons, particularly for NSAID and antibiotic compounds.

Due to the widespread use of WWTPs effluent in Saudi Arabia, the possibility of pharmaceuticals migrating from the unsaturated zone (soil) and contaminating groundwater was investigated using laboratory-scale soil column experiments. Most of the investigated pharmaceuticals had a high affinity for soil particles and tended to accumulate in the top 5 cm of the soil column. Therefore, no migration of these compounds to groundwater is anticipated in the natural environment, except for caffeine and cephalexin.

The estimated environmental risk from pharmaceutical compounds in the studied effluents was mostly medium, except for antibiotic compounds. Thus, antibiotics should be considered for future monitoring programes of the aquatic environment in Saudi Arabia.

P26 - Multivariate statistics and Machine Learning Tools to support the identification of chemicals in Baltic Sea sediments

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Chemicals have improved our quality life, but when released they can reach surface waters and enter the environment. Sediments are a sink for a myriad of organic chemicals and can also represent a significant risk for benthic organisms and coastal ecosystems, including consumption of fish species, as sediments become themselves a source of pollution if remobilized into the water phase.

Environmental samples are complex mixtures containing thousands of chemicals of anthropogenic sources and naturally produced, rending the identification of toxicants challenging. Bioassays can detect different types of effects but cannot identify the responsible chemicals. In this context, linking biological responses to chemical structures is demanding. New approaches, based on mass spectrometry (MS), were developed to prioritise the chemicals on which identification efforts can be made. Amongst them, virtual effect-directed analysis (v-EDA), was applied in water samples[1,2] and soils[3]. Another approach, developed in the last couple of years, is machine learning-based toxicity prediction tools like MS2Tox[4] and MLinvitroTox[5].

In this work, we collected 62 sediment samples along the Swedish Baltic shore (in the proximity or not of industrial activities) and in the open Sea. The chemicals analyses were performed by LC-MS in positive and negative mode. After peak picking, alignment, and clean- up, in total 90 000 and 30 000 m/z in positive and negative mode, respectively, were obtained (with about 10 000 MS/MS spectra available in each mode). Microtoxicity assays were performed, and dose response curved generated. As little differences were observed between the samples, EC_{10} were calculated. The chance to identify the toxicants explaining the microtoxicity response is almost impossible without the help of these prioritization approaches. *Our aim is to use the v-EDA and the machine learning approaches to investigate their applicability in projects containing many complex samples, resulting in large data sets, for future implementation.*

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P27 - Adaptation of Large Panels of Per- and Polyfluoronated Alkyl Substances (PFAS) for Routine Analysis of Drinking and Environmental Waters by Direct Injection Using UHPLC-MS/MS

Jenny Davies

Waters Corporatio

P28 - Utilizing ion mobility to enhance targeted and non-targeted analysis of PFAS from environmental samples collected as a ski resort

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P29 - TKIdentification platform for combined high-throughput Effect-Directed Analysis for the assessment of occurrence, fate and activity of Tyrosine Kinase Inhibitors as a novel class of pharmaceutical residues in the Dutch water cycle

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Routine monitoring of drinking water sources for pharmaceutical residues is commonly limited to the determination of known priority substances through chemical analysis. With the ever changing composition of the mixtures of compounds in the environment, other potentially hazardous chemicals (i.e. novel chemicals of emerging concern (CEC)) remain undetected. Tyrosine kinase inhibitors (TKIs) present a novel class of cytostatic contaminants in the water cycle. Despite potential adverse effects on ecosystem and public health, the impact of TKI residues has not been investigated. Biologically active metabolites and breakdown or transformation products can significantly contribute to the toxic potency of TKIs but remain undetected in target analysis. The advent of high-throughput effect-directed analysis (HT-EDA) allowed increasingly accurate and faster effect-based identification of bioactive chemicals in mixtures by combining high-resolution microfractionation and high-throughput screening on miniaturized bioassays together with analytical identification methods. However, current HT-EDA methodology lacks *in vitro* bioassays for kinase inhibition compatible with complex environmental mixtures. To achieve high-throughput detection and rapid identification of TKIs, bioassays are being developed and integrated with the previously developed HT-EDA platform.[1] Combined with enhanced non-target screening and recently developed workflows[2] the extended platform will be applied to the analysis of surface, drinking, and wastewater samples. Mixture of TKIs identified in samples will be tested in C. elegans and D. rerio to determine the combined toxic potency of environmental TKIs. This application of novel TKI bioassays with the HT-EDA platform will serve as blueprint for future expansion of HT-EDA for the assessment of currently relevant contaminants and to meet any future CEC challenges.

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P30 - In vitro toxicity profiling of various consumer products and toys

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- *In vitro* toxic potencies of various plastic products were evaluated on a tailored-set of CALUX bioassays e.g., for cytotoxicity (Cytotox CALUX), oxidative stress (Nrf2 CALUX), stable and lable AhR ligands (PAH and DR CALUX) and thyroid hormone disruption (TTR-TRβ CALUX).
- In this global survey (26 different countries from Africa, America, Asia and Europe) dioxin- and thyroid hormone-like activities were tested by CALUX and chemical analysis (BFRs) in consumer products and toys.
- Here we measured high levels of PBDD/Fs in these black plastic parts by using DR CALUX (from 110 to 17,000 pg BEQ/g) and by GC-HRMS (levels up to 13,900 pg TEQ/g).
- Also high TBBPA levels were measured by using TTR-TRβ CALUX (up to 410 μg/g) and by chemical analysis (up to 836 μg/g) indicating thyroid hormone disruption activities.
- The results indicate that a combination of effect-based bioanalysis CALUX bioassays and targeted congener specific chemical analysis would be a safer & more sustainable strategy to improve the assessment of the whole complex mixture of known and yet unknown additives and polymers from various plastic materials.

Reference: Behnisch et al. 2023. Environment International, 108079. https://doi.org/10.1016/j.envint.2023.108079

P31 - In vitro toxicity profiling of 45 PFAS compounds, technical products and water testing

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- *In vitro* toxic potencies of 41 PFAS compounds and one technical product (ADONA) were evaluated on a tailored-set of 6 CALUX bioassays for cytotoxicity (Cytotox CALUX), oxidative stress (Nrf2 CALUX), obesity/fat metabolism (PPARα/anti-PPARγ CALUX) and thyroid hormone disruption (anti-TRβ and TTR-TRβ CALUX).
- In vitro based relative potency factors (RPFs) are presented. The TTR-TRβ CALUX (disruption of TTR-T4 binding) was found to be the most responsive bioassay.
- For twenty surface waters and nine WWTP influent/effluent samples our PFAS reporter gene assay shows a good correlation to the chemical PFAS-13 analysis (both in PFOA-equivalents), albeit that the concentrations of the waste water samples found using the PFAS reporter are significantly higher than the chemical results. This indicates that in such waste water more unknown PFOA-like in vitro toxicity activities and compounds additional to the only 13 PFAS compounds here measured play a significant role for their thyroid transport hormone disruption.
- For the converted chemical analysis results this is comparable to the 5% percentile value of our study ranging here between 3.9 ng PFOA-eq./l (surface water) and 9.4 ng PFOA-eq./l (wastewater). In case of the bioanalysis results by PFAS reporter gene assays the 5%-percentile value is much higher with values of 560 ng PFOA-eq./l (surface water) and 710 ng PFOA-eq./l (wastewater), indicating that additional unknown PFAS may play a significant role in this in vitro thyroid toxicity pathway.
- The results indicate that a combination of effect-based analysis using e.g., the TTR- TRβ CALUX reporter gene bioassay and targeted congener specific chemical analysis would be a suitable strategy to improve the assessment of the whole complex mixture of known and yet unknown PFAS and their transformation products in the environment, wildlife and for public health.

Acknowledgments

The *in vitro* toxicity testing and resulting RPF values are a product of the PROMISCES Project, which runs from November 2021 to May 2025 and is funded by European Union’s Horizon 2020 research and innovation program under Grant Agreement No 101036449.

P32 - Identification of Compounds with Transthyretin-Binding Activity through Effect- Directed Analysis in Water Samples from the Upper Rhine

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Persistent, mobile, and toxic compounds (PMTs) have become a serious global threat to drinking water resources in recent years. These compounds, characterized by their polar, novel and often unknown chemical structures, can pose substantial risks to both human health and aquatic ecosystems. To address this challenge, effect-directed analysis (EDA) was employed to identify emerging PMTs.

This study shows the application of the EDA method for the analysis of surface and well water samples from the Rhine River. A sample preparation technique based on multi-layered solid-phase extraction capable of concentrating a wide range of PM compounds was applied. The prioritization of features for the identification was based on a transthyretin-binding assay [1] that measures the competitive binding of chemicals to the thyroid hormone distributor protein transthyretin (TTR). To enhance the identification process, a comprehensive database and suspect list of compounds exhibiting TTR-binding activity were established. This enabled the upgrade of the EDA data processing workflow [2], facilitating the detection of PM and other potentially hazardous compounds.

As a result, chemicals belonging to pharmaceuticals, industrial chemicals, surfactants, PFAS, and naturally occurring compounds were identified and confirmed on the Schymanski level 1. Most of the identified compounds demonstrated moderate to high potency in the TTR- binding assay.

Overall, this study highlights the effectiveness of EDA combined with the TTR-binding assay in identifying and assessing the risks associated with emerging compounds in water. The findings underscore the urgent need for proactive monitoring, targeted remediation strategies, and improved regulatory measures to safeguard human health and preserve water quality.

*Acknowledgmen*t – This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101036756.

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P33 - Categorisation of Mass Spectrometry data on the Basis of Toxicity

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Checking the quality of water, and especially drinking water, is becoming increasingly important, including the assessment of the toxicity of compounds (Samanipour et al., 2022). One of the challenges for the analysis of water is that a lot of compounds are still unknown and thus not present in libraries. Resulting in the need to annotate unknown compounds, and ultimately increase the time needed to perform assessment. When toxicity data is unknown, in vivo bioassays need to be performed, which are labour expensive. A recent study has shown that 3D structure of molecules expressed via molecular descriptors and fingerprints are able to explain the toxicity of chemicals. With the use of Random Forest and k-means a classification model made to classify the toxicity. This method might be suitable for toxicity assessment and may speed up the labour intensive process. For structurally known chemicals fingerprints provide enough structural information to correlate the presence of functional groups to the chemical's biological activities. However, since a lot of research is being conducted through untargeted analysis, most structures are not (fully) identified which makes the use of fingerprints impossible. In Figure 1the best k-means model for the fish toxicity is shown.

Another study showed that the application of cumulative neutral loss (CNL) data could be used to predict the RP-LC retention indices of structurally unknown chemicals from mass spectrometry (MS) data (Boelrijk et al., 2022). CNL is the difference between precursor ion and the fragments masses. For this reason, the classification model is built to categorise based on toxicity using CNL data. As mentioned earlier the model based on the fingerprints is made using Random Forest and labelled using k-means. To make the proposed model using CNL data more comparable, the same tools are used. The developed model is able to asses toxicity of features without prior annotation.

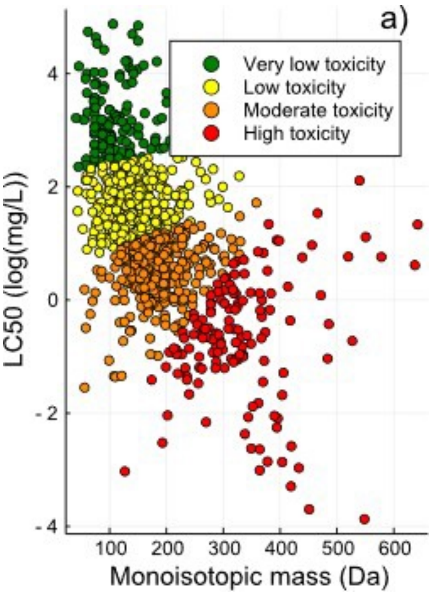

Figure 1: Distribution of the toxicity categories of the acute fish toxicity data set via the best k-means clustering model

Boelrijk, J., Herwerden, D. van, Samanipour, S., Ensing, B., & Forré, P. (2022). Predicting RP-LC retention indices of structurally unknown chemicals from mass spectrometry data. ChemRxiv. https://doi.org/10.26434/chemrxiv-2022-85wcl-v2

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P34 - Exposure and risk assessment of non-persistent pesticide biomarkers in Spanish adult population using human biomonitoring

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Text: Six non-specific pesticide metabolites, 21 specific pesticide metabolites and 13 pesticides were studied in 380 urine samples provided from adult population (aged 18-65 years old) residing in the Valencian Region, Spain. The different employed analytical strategies, including dilute and shoot and micro-solid phase extraction (µSPE) enable us to use less than 1 mL to perform the analysis by LC-MS/MS. 36 out of 40 assessed biomarkers were detected at least one sample, with detection frequencies ranged from 1% to 95 %. The detected biomarkers with the highest detection frequency were DEET, Boscalid-OH, Tebuconazole-OH, PNP, PBA and DEAMPY, whose geometric mean concentrations ranged from 1.28 ng μL^{-1} to 4.64 ng μL^{-1}. Estimated daily intake (EDI) and hazard quotient (HQ) were obtained in order to interpret urinary levels of the most frequently detected pesticide metabolites in a risk assessment context. The calculated HQs were lower than 1, showing that a low health risk due to the pesticide exposure was observed.

P35 - Heavy Metal Contamination in Food by ICP-MS method: Occurrence and Human Health Risk Assessment in Cyprus

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An accredited method using inductively coupled plasma mass spectrometry was applied for the determination of lead, cadmium and mercury in several food categories such as fish and mammals' meat, molluscs, milk, honey, fruits and vegetables, grains and grain products and foods for infants and children. High percentage of the samples were contaminated with at least one of the three heavy metals with concentrations ranging between 0.0002 and 0.379 mg kg^{-1}. The dietary exposure assessment to the heavy metals, lead, cadmium and mercury, was performed using the deterministic risk assessment tool, ImproRisk. ImproRisk estimates the dietary exposure at individual level by combining individual consumption data and occurrence data, which are both coded according to the EFSA's food classification and description system, FoodEx 2. No health risk was concluded from the dietary exposure assessment to lead, and also less than 1 % of the Cyprus population was found to exceed the health-based guidance value (Tolerable Weekly Intake, TWI), as regards to mercury. On the other hand, a significant percentage of infants and children exceeded the TWI for cadmium intake (Figure 1). The dietary exposure to heavy metals was found to be independent of the geographical area in Cyprus and gender. Exposure to lead was uniformly distributed to all food categories, while molluscs and potatoes were the highest contributors to the cadmium intake. In the case of mercury, fish, meat and milk contributed the most to the total exposure of the population in Cyprus.

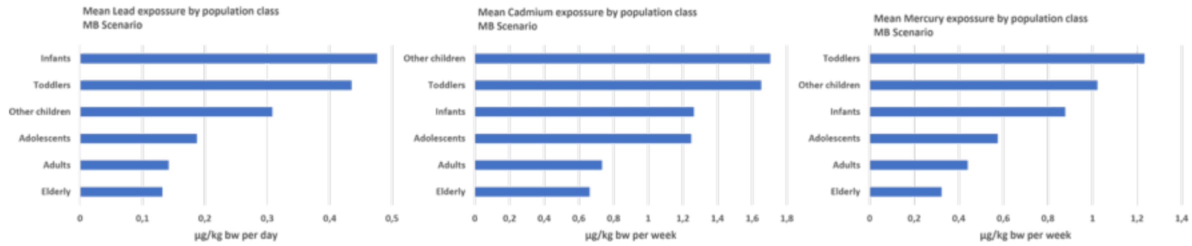

Figure 1. Dietary exposure to heavy metals by population class.

P36 - Levels of thirteen toxic elements in blood of adult population of Valencia Region (Spain)

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Population is exposed to toxic elements through ingestión, inhalation and dermal contact mainly. Exposure of high levels of these contaminants has been associated with a high number of health effects such as cancer, neurologic disorders or cardiovascular diseases. Biomonitoring allows to assess the internal exposure to these contaminants using human matrices such as blood, hair or urine. [1]

In the present study, thirteen toxic elements were determined in 496 blood samples from adult population of Valencia Region (Spain). Samples were collected during 2021 in the context of the BIOMOVAL project. Hg was measured using a direct mercury analyzer while the rest of coumpounds were determined using an ICP mass spectrometer.

Five (Hg, As, Sb, Cs and Pb) out of thirteen elements showed high detection frequencies (>90%), Al detection frequency was 22% and the rest of elements detection frequencies were low(<10%). The concentrations ranged from <1.0 µg/L to 521µg/L, and Pb showed the highest concentration with a geometric mean of 12.1 µg/L.

Levels of Hg, As and Sb were higher than in previous studies implemented in adult population in other countries, while Pb levels were in general lower than in previous studies. In a risk assessment context, 41% of the participants of the present study had higher concentrations of Hg than the HBM-I guidance value (5 µg/L), and 7% showed levels higher than the HBM-II guidance value (15 µg/L) [2]. Therefore, measures should be taken to decrease Hg exposure in the studied population.

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P37 - Biomonitoring of essential metals in blood of adult population from Valencian Region (Spain)

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Levels of seven essential metals were evaluated in blood of 496 adult participants of the BIOMOVAL program, a human biomonitoring study carried out in the Valencian Region (Spain). The method of analysis consists of a 1:20 dilution with 0.5 mL of sample, to which 9.4 mL of H_2O type I, 50 µL of 0.5% Triton X-100 and 50 µL of 69% HNO_3 are added, with subsequent measurement of the analytes in an ICP-MS. The seven essential elements analyzed were Co, Cu, Mn, Mo, Se, V and Zn and the LoQ was 1 µg/L for all of them, except for Cu and Zn which was 20 µg/L. Co, Mo and V presented a detection frequency below 40%, and for the rest of analytes was 100%, being the geometric mean 847 µg/L for Cu, 7.0 µg/L for Mn, 168 µg/L for Se and 5582 µg/L for Zn. In comparison with concentrations found in previous studies in adult populations of other countries, Mo, Co and V also showed values lower than 1 µg/L in studies implemented in Germany[1], the USA[2] or Canada[3]. As for the rest of the essential elements that could be quantified, all the levels obtained were lower than those reported in these studies, except for Se. For instance, Zn showed a value of 5620 µg/L in Germany and 5600 µg/L in Canada; Cu 934 µg/L in Germany and 980 µg/L in Canada; and Mn 8.5 µg/L in Germany, 9.5 µg/L in the USA and 10 µg/L in Canada. In the case of Se, the population of the Valencian Region showed a higher concentration than Germany (106 µg/L) but lower than Canada and the USA (170 µg/L and 188 µg/L, respectively).

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P38 - Climate Change and Food – Effects of Increased CO_2 Concentrations and Drought Stress on the Metabolic Profile of Soybeans

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Plants are continuously exposed to rising CO_2 levels, as the concentration in the atmosphere increases by 1.5-2.0 ppm annually. At the same time, more and more plants are experiencing further challenges from water shortages. The change in these living conditions requires corresponding physiological adaptations from most organisms.

Although science and politics have been tracking the effects of such exogenous factors for more than three decades, it is not yet clear what influence climate change will have on the composition of our food in the long term. The results of previous studies are not always unambiguous in this respect and seem to depend strongly on the plant genera or species considered, with sometimes contradictory results[1].

Considering this diffuse data situation, soybean plants were cultivated under different CO_2 and water contents in the present study. Leaves were examined at regular intervals to analyze gas exchange. In addition, the metabolic profile of the soybeans was analyzed using high- resolution mass spectrometry (LC-ESI-IM-QTOF, non-targeted metabolomics) and then evaluated by means of various multivariate evaluation strategies.

The gas exchange measurements have shown that the CO_2 uptake rate increases under elevated CO_2 concentrations and decreases under drought stress. The rate of transpiration decreases under drought stress, but does not respond to increased CO_2 concentration in the atmosphere. While the drought stress conditions did not have strong effects on the metabolic profile of soybeans, numerous differences in the metabolome could be observed in the plants cultivated at different CO_2 levels. These concerned, in particular, the lipid composition of soybeans.

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P39 - An analytical strategy for assessing the absence of titania particles in food products on the EU market following the E 171 ban as a food additive

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Food-grade titanium dioxide, known as E 171 in the EU, is a widely used food additive that, owing to the light-scattering effect of TiO_2 particles occurring in the particle size range 200– 300 nm, is used as a whitener. It is a polydisperse material composed by anatase (rarely rutile) particles, with a constituent size typically ranging from 30 to 350 nm. Following the safety re-assessment of the European Food Safety Authority (EFSA) in 2021, E 171 has been banned as a food additive in the European Union (EU) in 2022.

Testing compliance of food products on the EU market with the E 171 ban poses significant challenges to official control laboratories. TEM or SEM imaging coupled with elemental analysis (e.g. EDX) represents the confirmatory approach providing certainty of E 171 presence along with the capability to characterize the size of the constituent particles and their number-based size distribution. Over the last years, single-particle ICP-MS (spICP-MS) has emerged as an ideal technique for E 171 analysis with widespread availability in testing laboratories, higher sample throughput, and lower analytical costs. However, there is a need for efficient screening methods to identify samples to be tested by this particle- and elemental- specific method. Determination of total titanium by ICP-MS is a practicable option and was investigated in the present study. The premise of such an approach are the relatively low and uniform Ti concentrations in food, unless manmade TiO_2 food additives are added. In addition to E 171, this encompasses pearlescent pigments consisting of mica platelets coated with TiO_2 nanoparticles, whose use is also illegal.

Herein we describe the analytical strategy we developed for assessing the absence of titania particles in food products on the EU market, founded on total Ti determination via ICP-MS as screening method, followed by spICP-MS analysis as confirmatory method. A tailored sample preparation, i.e. a microwave digestion method resulting in solubilization of TiO_2 particles for total Ti, and a chemical extraction procedure of TiO_2 particles for spICP-MS analysis, was set up.

P40 - Human and farm animal exposure to pesticides - Silicone wristbands to study non-dietary routes of exposure

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Silicone wristbands have emerged as a suitable passive sampler for evaluating pesticide exposures in both humans and animals. Until now they have been deployed to assess a limited number of pesticides across some species.

We measured 177 pesticides (including some metabolites) in wristbands (2 to 5 gram each) worn by 715 humans, 152 farm-animals (including 21 farm cats), across 10 countries in Europe and Argentina, in close proximity to both conventional and organic farming activities. The extracts from these were analyzed via LC-MS/MS. For humans, we collected information on possible exposure determinants, such as frequency of house cleaning, time spent indoors, use of pesticides. Multivariable generalized least squares (GLS) models were used to study determinants of wristband concentrations.

Overall we detected 171 out of 177 pesticides targeted in our analysis. A mixture of fludioxonil, boscalid, fipronil, azoxystrobin and piperonyl butoxide was detected in 30% of the human wristbands. In animals, pendimethalin paired with piperonyl butoxide in >30% wristbands. The average concentrations ranged from 0.5 to 117 ng/g in humans and 0.2 to 487 ng/g in farm-animals and cats. Maximum concentrations measured were 5,500 and 14,062 ng/g for dimethomorph in humans and propoxur in farm-animals, respectively.

Concentrations in wristbands worn from participants living close to conventional farms were higher in comparison to those living close to organic farms. Having pets increased significantly the fipronil and fipronil sulfone concentrations. For other pesticides, reporting spraying or working in agricultural sector turned out to be the main predictors of elevated wrist band concentrations.

Wristbands proved to be a good proxy for environmental exposure assessment. Multiple pesticides were captured in both wristbands worn by humans and animals, which allowed to look at exposure contrasts within and between species. In future studies, wristbands can be used to prioritize in internal exposure studies (human biomonitoring), and to study links between environmental exposure and disease development across species and for multiple compounds.

P41 - Monitoring of pesticides in human and animal feces: method, validation and application

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Exposure assessment of pesticides is traditionally based on residues measured in food, combined with food consumption data. Biomonitoring is an alternative approach, where pesticides and/or metabolites are measured in body fluids or other biological matrices to assess internal exposure. Urine is the most common matrix analyzed, followed by blood. Blood has the disadvantage of being an invasive matrix complicating sample collection, and reflects only a very short exposure window. In urine, pesticides are typically excreted as their metabolites which complicates their determination. In addition, for some pesticides urinary excretion is only a minor route. Feces so far has hardly been explored for biomonitoring. In this work, for the first time, multi-residue methods were developed and validated for the determination 205 pesticides and metabolites in fecal samples of humans and animals (cattle, sheep, pig, chicken, cat, bat). Applying the QuEChERS extraction protocol, the method was evaluated at 1, 2.5, 5 and 10 µg kg^{-1}, using both LC and GC-MS/MS techniques. Almost 70% of the compounds presented acceptable recoveries and relative standard deviation at the lowest level tested. For LC compounds substantial signal suppression was observed thus matrix matched standards were required for quantification

purposes. The method was applied for the analysis of approximately 700 samples of human feces and 250 samples of animal feces from 10 EU countries and Argentina within the SPRINT project (https://sprint-h2020.eu/). Overall, 94 pesticides were detected, typically in the 1-10 µg/kg range, but also 10-100 µg/kg or even higher. Pyrethroids, organochlorine pesticides, and fipronil-sulfone were most frequently detected. This work shows that fecal analysis provides additional exposure information.

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P42 - Pharmaceuticals in municipal WWTPs in Poland in the aspect of different agglomerations

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Abstract: *The presence of pharmaceuticals in the aquatic environment is known to scientists* and is constantly being investigated. Such micropollutants with bactericidal, virucidal, or fungicidal properties are commonly used in households, which results in their presence in raw sewage flowing to wastewater treatment plants. The content of pharmaceuticals may vary depending on the lifestyle of the inhabitants or the intensity of drug consumption. This study analysed the content of antibiotics, virucidal and fungicidal substances in raw and treated sewage from two cities with a population equivalent of 68,000 and 250,000 located in southern Poland. Furthermore, the study was conducted twice, ie, in the summer and autumn-winter seasons.

The purpose of the study was to compare the concentrations of pharmaceuticals in the sewage of two cities that differ in the amount of sewage and the number of inhabitants and to determine the degree of reduction of marked micropollutants in the treated sewage. Analysis was performed using SPE extraction and UPLC-MS/MS chromatography. An attempt was made to determine 51 different compounds that are bacteriostatic, fungicidal, and virucidal drugs and their derivatives in average daily sewage samples.

The presence of 33 pharmaceuticals was detected and 16 of them were determined in the wastewater of a smaller city. In the sewage of a larger city, 42 substances were detected and *27 of them were marked. During the wastewater treatment process, the content of* pharmaceuticals in the wastewater was reduced (e.g., sulfasalazine, erythromycin, sulfapyridine N-acetyl), as evidenced by the reduction of the concentrations of the analysed substances in almost 100% of the cases.

Studies have shown the presence of more pharmaceuticals and their higher concentration in city municipal wastewater with a higher population equivalent value. A decrease in drug concentration was observed in both wastewater treatment plants, with the level of reduction being greater in samples taken in the summer.

Keywords: *pharmaceuticals, micropollutants, sewage, wastewater treatment plant*

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P43 - Screening of 850 pesticides in soil, water and food of plant origin using LC-Q-Orbitrap

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Abstract:

We present an approach for the screening of pesticide residues and their metabolites in soil, water and food on a Thermo LC-Q-Orbitrap instrument (Q-Exactive classic). We used the Thermo Scientific Environmental and Food Safety HRAM compound database with 1700 entries, as well as the corresponding EFS HRAM spectral library as starting points to build an in-house compound database (Tracefinder™) and spectral library (mzVault™) of 850 pesticides, metabolites and co-formulants. We have reference standards of 450 of the compounds, with known retention times.

There have been discussions on whether the data-independent fragment acquisition mode (DIA) or the data-dependent fragmentation (ddMS2) is the more optimum for screening methods. However, we find that a combination of both provides the best approach, using FullMS-DIA for non-targeted screening and FullMS-ddMS2 for targeted screening and confirmation.

The Thermo Scientific Accucore aQ column, in combination with the UltiMate 3000 UHPLC and QExactive instrument has proven a very robust setup for the screening of pesticides. The retention times remain stable for years and sensitivity is very good, achieving LOQs of 10 µg/kg or better for most compounds. Matrix enhancement or suppression effects, which often are a problem in triple quadrupole instruments, are negligible in samples run on our QExactive instrument so far. We participate annually in screening proficiency tests that verify our method performance in detecting both known and unknown compounds.

We use the method to screen for pesticides and metabolites in soil and water samples in Norway, as well as in domestic and imported food. As a national reference laboratory for the analysis of pesticide residues in food we also have sensitive targeted multimethods on triple quadrupole instruments, but the screening method on high-resolution MS is superior in detecting new contaminants not included in the scope of the triple quadrupole methods. The screening method also provides us the option to detect compounds in a single run, that otherwise would have to be analyzed both with LC-MS/MS and GC-MS/MS in our monitoring routine scope.

P44 - RISK ASSESSMENT OF SEVERAL ORGANIC MATERIALS IN CONTACT WITH WATER FOR HUMAN CONSUMPTION

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Water intended for human consumption comes into contact with different construction materials used in collection, treatment, storage and distribution systems up to the consumer's tap. All materials used in water supply systems can have an impact on the quality of drinking water through the migration of potentially harmful substances, by enhancing microbial growth or by influencing the odour, colour or taste of such water [1]. There are several European Standards with the methodologies for testing and accepting final materials.

The Analytical Water Laboratory of EPAL performs and is accredited for seven European standards related to migration tests on organic materials and on cementitious materials. All analytical tests carried out in the migration waters are also accredited (metals, pH, conductivity, organoleptic parameters, and identification of unknown organic compounds by GC-MS).

In this study, the risk assessment of eighteen organic materials tested in the laboratory, from ten different manufacturers, over the past two years was performed through the potential for non-target compounds from the materials to migrate into the water. The organic materials based on polyurethane, polyurea and EPDM rubber were the ones with the highest number of organic compounds identified in the migration tests using a GC-MS analytical method. Among the 133 different organic compounds detected, the highest percentage were non-intentionally added substances (85% of total), and only 15% of the identified substances are listed on the European positive lists (EUPLs) of starting substances for use in manufacture of organic materials in contact with water intended for human consumption. The manufacturing process of PE pipes was also evaluated through the following variables: use of raw materials from different suppliers for the production of PE pipes in the same company; and use of the same raw material for the production of PE pipes in different companies.

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P45 - Modular open-access and open-source Julia HRMS toolbox

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Non-target analysis (NTA) of biological or environmental samples analyzed with chromatography coupled with high-resolution mass spectrometry (HRMS), generally yields complex data. While there are a variety of tools and software available for the processing of these complex datasets, not all programs are open-source or open-access, provide sufficient freedom to the user to tailor their data processing to the needs of the research question, or are fully suited for the processing of NTA data as they may be more tailored to targeted or suspect screening approaches. Therefore, we developed a modular open-source and open-access framework for the processing of HRMS data.

The HRMS toolbox allows the user to perform a variety of different workflows, containing steps from feature detection to trend analysis. The steps related to processing the raw data, which can be performed on variety of analysis methods (e.g., data independent analysis and data dependent analysis), are feature detection (SAFD), componentization (CompCreate), and suspect screening. Then, for the interpretation of the extracted information, library searching (ULSA), alignment of features or components, and statistical analysis (i.e., PLS-R, PLS-DA, HCA, SVD, and k-means) could be performed. The library search algorithm comes with a built-in database that contains spectra from MassBank EU, MassBank of North America, and the Global Natural Products Social Molecular Networking (GNPS). Alternatively, the user can provide their own database given that it is formatted correctly. Moreover, the HRMS toolbox allows significant modularity, which enables users to perform steps within a workflow using different algorithms or programs and use this output within the HRMS toolbox. For example, the output of Patroon can be easily converted to that of the HRMS toolbox and vice-versa. Additionally, the toolbox comes with a set of built-in visualization tools that enable the user to evaluate their raw data and the results from the different algorithms. Finally, the toolbox also allows users to perform a variety of statistical analysis approaches for initial screening to evaluate if these could provide useful insight or trends in a dataset.

Overall, the HRMS toolbox is an open-access and open-source program developed for the analysis of HRMS data, which can be used in a highly modular way as well as being tailored to user specific needs. To showcase the toolbox, a set of wastewater and biological samples has been processed.

P46 - Spectral quality scoring

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The liquid chromatography-high-resolution mass spectrometry (LC-HRMS) combined with untargeted analysis has been applied in different research areas such as environmental analysis, metabolomics, forensics, etc. These assays are performed in data-dependent (DDA) and data-independent (DIA) acquisition modes, depending on the application area. While data acquired in DDA mode is inherently biased towards highly abundant features or predefined precursor ions [1], DIA is an unbiased approach that enables the acquisition of fragmentation spectra for all detected precursor ions, thus providing holistic information about the constituents of the sample. Since a large number of precursor ions are fragmented simultaneously, DIA generates highly complex data, the quality of which is extremely important for robust and reliable data processing.

This study focuses on optimizing LC-HRMS experiments using both DDA and DIA modes. Traditionally, optimization is conducted based on identification through spectral matching, often disregarding thorough investigation of data quality. In this study, MS2 data quality serves as a benchmark for optimization. Considering that DDA generally produces higher-quality spectra [2], this work compares spectra acquired in DDA and DIA modes to establish a quality score for spectral quality assessment.

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P47 - Biofilm as an innovative passive sampler for characterizing wastewaters by NTS and in vitro bioassays

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Last decade, non-target screening (NTS) analysis has been increasingly used to detect a large range of micropollutants in water, including ones of emerging concern, which positions it as a great tool for environmental monitoring (Schymanski et al., 2014). In parallel, passive samplers (PS) are often considered for this purpose due to their ability to accumulate substances that are not concentrated enough to exceed the detection limits of analytical instruments when classical sampling methods are used (e.g., grab or composite sampling), and because they offer great time-representativeness. In addition, combination of PS and bioassays has already shown its effectiveness to characterize a large range of compounds (Ahkola et al., 2021). However, this sampling technique is scarcely used in wastewater due to the complexity of this matrix. Recent studies have demonstrated the potential of aquatic natural biofilms to bioaccumulate micropollutants: biofilms have therefore been used as passive samplers, especially in wastewater, in which they can grow easily and quickly (Aydin et al., 2020; Huerta et al., 2016). The aim of this work was therefore to optimize the extraction step of these biofilms for NTS purposes and bioassays to ensure the largest range of extracted compounds and to evaluate its added value compared to classical sampling methods. Thus, several solvents and extraction methods were compared for biofilm extraction, while wastewater samples were extracted using an already optimized technique that enables the coverage of a large range of micropollutants (i.e., multilayered SPE cartridge) (Huynh et al., 2021). The evaluation of the extraction efficiency was based on high-resolution mass spectrometry characteristics such as standard recoveries, number of common and specific compounds detected with suspect screening, number of common and specific unknown features detected, range of molecular weight or polarity. First results demonstrate that biofilms can accumulate a large range of organic micropollutants from wastewaters and the comparison of grab samples analysis demonstrated differences in fingerprint pattern. Bioassays were also applied to evaluate the relevance of this innovative sampling method to assess the endocrine disrupting effects of the micropollutants present in the wastewaters.

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P48 - Improved target, suspect- and non-target analysis of environmental contaminants in wastewater using a GC-EI&CI-TOF-MS system

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Wastewater plants can release aqueous effluents containing harmful pollutants, posing risks to ecosystems and human health. Given the dynamic nature of wastewater composition, regular target and non-target analyses are frequently conducted. Due to its complimentary polarity range, GC-MS is often employed alongside LC-ESI-HRMS to evaluate potential hazard profiles and to identify compounds in these complex mixtures and to gain a more complete overview of the potential hazards present in the wastewater. However, conventional GC-EI-MS is limited in its identification confidence for some compounds due to unspecific fragmentation pattern, absent molecular ion signals or by not being available in reference libraries, leading to a lack of compound identification.

The ecTOF (TOFWERK, Switzerland) is a newly developed dual ionization source time-of-flight (TOF) mass spectrometer. The instrument consists of a standard TOF analyzer operating two ionization sources quasi-simultaneously: a standard 70 eV EI source and a medium pressure CI source (HRP; TOFWERK, Switzerland) (Figure 1). For this study, 24h effluent samples were collected from an industrial plant over two months. Analytes were enriched by headspace (HS) solid-phase microextraction (SPME) using a novel hydrophilic-lipophilic balanced (HLB) fiber coating. An Agilent 7890A gas chromatograph (Agilent, Santa Clara CA, USA) was coupled to the ecTOF.

This talk will highlight the advantages of the simultaneous acquisition of EI and CI mass spectra for contaminant identification of complex environmental samples within one GC run. The tentative identification processes and statistical approaches that can be employed using the GC-ecTOF data will be shown. The simple selection of different CI reagents ions (e.g., $[NH_4]^+$, $[N_2H]^+$, $[H_3O]^+$) between chromatographic runs, which enables the adjustment of reactant selectivity and the degree of fragmentation, is proven to be highly valuable for the compound identification process. Furthermore, the quasi-simultaneously generated EI and CI mass spectra increased identification yields and reduced false positive rates compared to standard GC-MS methods. With this, the GC-ecTOF can be used to complement methods like HPLC-ESI-HRMS commonly used in industrial wastewater monitoring.

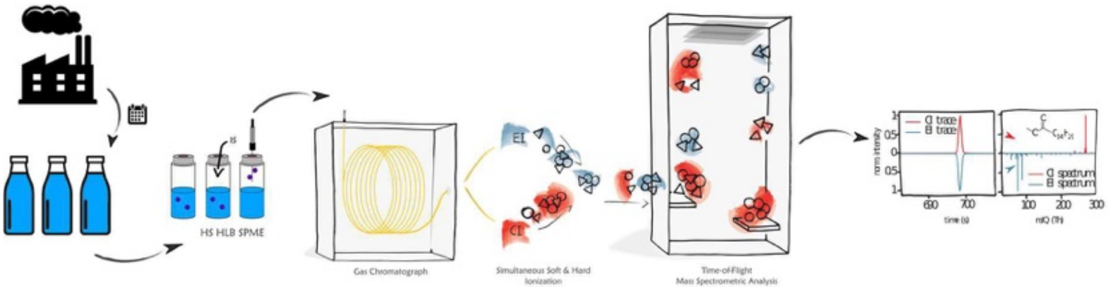

Figure 1: Schematic experimental setup for wastewater analysis using HS HLB SPME in combination with the GC-ecTOF. Example chromatogram and spectra of EI and CI data are also shown.

P49 - Improved compound identification in GC analysis using an EI&CI-TOFMS

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A time of flight (TOF) mass spectrometer simultaneously operating an electron ionization (EI) and a chemical ionization (CI) source is presented for non-target analysis. By coupling both ionization sources directly to one single gas chromatograph (GC), structural as well as accurate mass molecular ion information is generated simultaneously from each analyte. Hence, target and suspect screening analysis as well as effective non-target analysis using GC-MS is improved considerably. In this poster, various experiments will be discussed, illustrating the potential of the GC-ecTOF especially for non-targeted approaches, including applications in fields such as environmental contaminants, material emissions, food and flavor analysis and metabolomic research.

An Agilent 6890A GC was coupled to an ecTOF (TOFWERK, Thun, Switzerland), using a 70 eV EI source and the newly developed Tofwerk HRP CI source. Various GC methods and sampling procedures were employed depending on the analytical need of the study, including liquid injection of extracted samples, headspace sampling including SPME and thermal desorption using Tenax tubes. To generate the ideal molecular ion information different reactant ions (e.g., $[NH_4]^+$, $[N_2H]^+$, $[H_3O]^+$) were used for the chemical ionization process.

Standard procedures employed by routine laboratories, e.g., target screening for material emissions or steroid screening, are shown to be feasible using the GC-ecTOF. Whilst standard methods mainly focus on target analysis, suspect screening is improved and non-target analysis even enabled using the ecTOF. Especially when EI library hits are fair with low corresponding probability, chromatographic and CI information can be used to increase compound identification confidence. False positives from an EI only approach can easily be identified and often correctly annotated using the additional information from the ecTOF. Furthermore, compounds that are not listed in libraries have a high uncertainty for identification using an EI only approach. Using the accurate mass information generated by CI, molecular sum formula for these unknowns can be derived. Combining this with the structural information generated by EI, tentative structure elucidation becomes feasible in many cases. Hence, non-target approaches become viable using the GC-ecTOF.

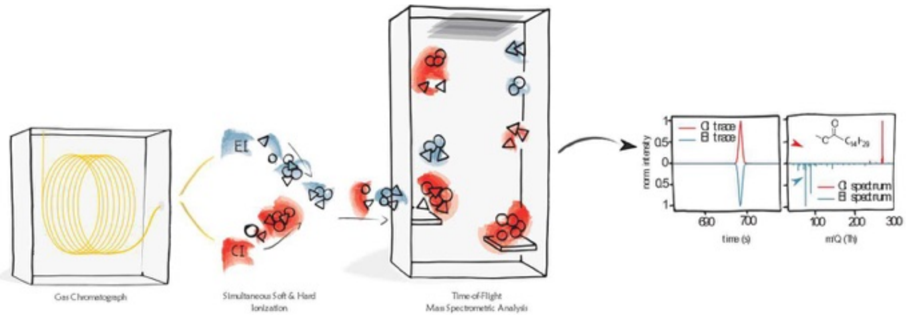

Figure 1: Principal operation for GC-ecTOF with an example chromatogram and spectra of EI and CI data.

P50 - Temporal and spatial patterns of xenobiotics in German fish

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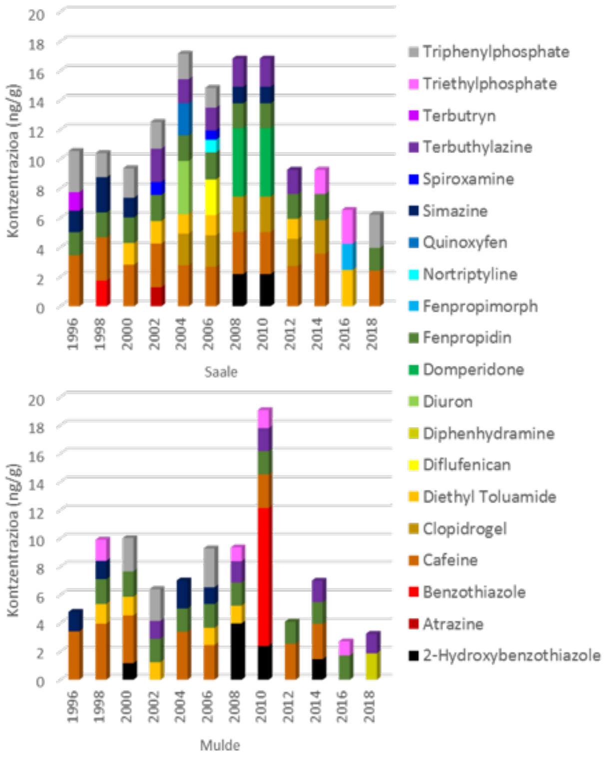
The complexity of the occurrence of emerging contaminants in aquatic environments is remarkable. The different physicochemical properties of the compounds and thus, their degradation and bioaccumulation processes affecting aquatic organisms are often missing due to a lack of analytical procedures to measure emerging compounds in biota. Moreover, the temporal trends in the occurrence of compounds that have been overlooked in the monitoring programs since decades are a scientific gap that could be tackled thanks to the sample repositories stored in the specimen banks across Europe.

The objective of this study is to apply the recently optimized protocol in the Plentzia Marine Station (EHU/UPV) to analyze xenobiotics in a biological tissue (fish filet) from a non-target point of view in historical and present biota samples.[1] A collection provided by the German Specimen Bank of 24 samples of bream from the Saale and Mulde rivers gathered biannually from 1996 to 2018, plus 25 samples from other rivers in Germany, including Rhine, Elba, Danube, and Saar among others, taken in 2018 was analyzed. The assessment was performed on a Thermo Scientific Dionex UltiMate 3000 UHPLC coupled to a Thermo Scientific™ Q Exactive™ Focus quadrupole- Orbitrap equipped with a heated ESI source (Thermo, CA, USA). Analysis of the extracts was performed in positive and negative mode in the Full scan data-dependent MS2 discovery acquisition. A selection of circa 250 compounds, including personal care products and pharmaceuticals, consumption products, industrial compounds (PFAS, plasticizers), and pesticides was used for the target analysis. Finally, a full suspect analysis with a database of more than 40,000 compounds from the NORMAN list will be presented.



Distribution of the top 20 contaminants in the period 1996 – 2018 in Mulde (down) and Saale (top) river fishes.

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P51 - Identification of an unknown compound in rapeseed using combined orbitrap and iontrap MS-analysis

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Rapeseed (*Brassica napus*) is used as animal feed for cattle, pigs and poultry. In multiple rapeseed samples, routine pesticide analysis using LC-MS/MS resulted in false positives for the herbicide metamitron. Both MRM transitions 203>175 and 203>104 were found at the retention time for metamitron. The ion ratio deviated 25% from the ion ratio in standard solution, which was within the 30% criterion in SANTE/11312/2021 [1]. However, in the sample with standard addition, the ion ratio deviation was only 9%. Therefore investigation into the identity of this compound was initiated.

A rapeseed sample was extracted as such and spiked with metamitron at 100 ppb level and analyzed on a Thermo Scientific IQ-X HRMS, using the same chromatographic conditions as in the original LC-MS/MS method. Full scan and data dependent MS2 (dd-MS2) orbitrap spectra were acquired. In the same measurement, dd-MS2 spectra were acquired in the ion trap, to obtain data similar to the original LC-MS/MS analysis.

Evaluation of the full scan orbitrap data revealed the presence of m/z 203.0815 in the rapeseed sample. Metamitron (m/z 203.0927) was only present in the spiked sample, confirming that the original LC-MS/MS result was a false positive. In the dd-MS^2 ion trap spectra, both metamitron and the unknown compound fragment to m/z 104.1 and 175.1 which corresponds to the LC-MS/MS results. The fragment m/z 104.0495 (C7H6N+) was present in the dd-MS^2 orbitrap spectra of both metamitron and the unknown compound. However the metamitron fragment m/z 175.0978 was not present in the spectra of the unknown compound, where m/z 175.0866 was detected instead. To obtain an annotation for the unknown compound, the data was evaluated using Freestyle (Thermo Scientific), MassFrontier (Thermo Scientific) and MSFinder [2] software. The molecular formula for the unknown compound was annotated as $C_{11}H_{10}N_2O_2$. A mzCloud search was performed but no match to a mzCloud library spectrum was made. In MSFinder a result based on in-silico fragmentation was obtained, identifying the unknown compound as the natural compound 1,2-dihydrospiro[indole-3,3'-pyrrolidine]-2,2'-dione.

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P52 - The importance of establishing a research consortium to harmonize data processing and *in-silico* tools in suspect and non-target screening studies: A call to action

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Large-scale suspect and non-targeted screening (NTS) approaches based on high-resolution mass spectrometry are being used more frequently in environmental monitoring and food safety[1,2]. Although much progress has been made in harmonizing workflows from an analytical perspective to ensure reproducibility, the same is not true of the downstream computational methods used to analyse the large amounts of data generated in these studies[1]. Data processing and analysis applied to NTS requires advanced computational tools—many of which are still in development. Additionally, the latest computational tools developed in other research fields, such as proteomics and metabolomics, are beginning to be implemented in environmental monitoring and food safety. But, due to the specific expertise required to analyse this highly complex data, it is common practice for each laboratory to use its own software and methods optimized for its own needs. However, what might be considered the best "in-house" approach to solving a particular problem may not necessarily be the best solution from the perspective of the wider research community. The methods, algorithms and software available to process and analyse data for a particular application are far from standardized and there is no harmonization between different laboratories working with similar problems. This can lead to spurious correlations, false discoveries, and loss of important information. Therefore, there is a critical need to harmonize these methods to obtain reproducible results, make data comparable, and join efforts to discover emerging risks before they become a major concern[3]. With this in mind, we suggest establishing a research consortium with the aim of harmonizing data processing and in-silico tools used in suspect and non-target screening studies. We believe that, analogous to the CASMI challenge (Critical Assessment of Small Molecule Identification) in the plant metabolomics community, a first step might be to share known datasets and metadata (derived from real suspect and non-targeted screening studies) among participants to jointly identify the best approaches and key parameters to ensure consolidated outputs, while maintaining the flexibility of the laboratories and their capacity in discovery and exploratory research. The establishment of international standards to follow in data processing and in-silico methods to flag, annotate and identify emerging contaminants should also be a key outcome of the research consortium's agenda. Such standards must be in line with the FAIR principles, so that the data, software and protocols are Findable, Accessible, Interoperable and Reusable to achieve better consolidation and comparability of data generated from various studies, especially in regards to its further use in a regulatory framework and in a policy support context.

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P53 - Uncovering the PFAS Complexity: A powerful LC-VIP HESI(-)-TIMS-QToF MS Workflow for Biota Analysis combining targeted and untargeted approaches

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With their widespread presence in the environment and organisms, and their persistent, bioaccumulative, and toxic (PBT) properties, Per- and Polyfluoroalkyl Substances (PFAS) pose a significant challenge as organic micropollutants. The vast array of commercially available PFAS compounds and their transformation products necessitate an intricate monitoring approach within environmental compartments.

This study aims to the development of a combined targeted and untargeted workflow for the thorough characterization of PFAS in complex environmental matrices, particularly biota, by incorporating trapped ion mobility spectrometry (TIMS) into LC-HRMS methodologies. Additionally, the study showcases the analytical performance of RPLC-VIP HESI(-)-TIMS- QTOF MS in environmental applications.

To enable untargeted screening, biota extracts were prepared using a generic protocol designed for the simultaneous extraction of 56 PFAS compounds from various sub-groups. RPLC-VIP HESI(-)-TIMS-QTOF MS was employed for analyzing the extracts. Targeted workflow utilized broadband collision-induced dissociation (bbCID) as a data-independent acquisition mode, while untargeted workflow employed PASEF as an efficient data-dependent mode. The CCS-aware target analysis incorporated a list of 60 PFAS compounds, including precursor ion formula, retention time, MS1 and MS2 qualifier ions, and ion mobility-derived collision cross section (CCS) values. The results of target screening demonstrated improved sensitivity and lower PFAS detection limits due to VIP-HESI, along with higher quality full-scan MS and bbCID MS/MS spectra enabled by ion mobility filtration. Furthermore, collisional cross sections (CCS) enhanced the identification confidence.

In the untargeted workflow, raw data was transformed into a comprehensive feature table, and the detected features were prioritized using Kendrick mass analysis. A PFAS suspect list comprising approximately 5,000 compounds was employed for further annotation. *In-silico* prediction of MS/MS spectra and CCS values aided in the identification of suspected compounds. Notable examples highlighting the prioritization and annotation of suspected features as PFAS-related compounds were identified, leveraging the extensive MS2 coverage provided by PASEF.

The proposed workflow combining LC-TIMS-HRMS with target, suspect, and non-target screening facilitates comprehensive PFAS monitoring. This approach contributes to a better understanding of the chemical universe of PFAS in the environment and plays a crucial role in safeguarding the environment, wildlife, and human health in a One Health approach.-Bruker Confidential-

P54 - Identifying unknown bacterial stressors in environmental water samples.

List of authors: J. García Martínez[1], M. Islam[2], Z. Mthiyane[3], M. Sandahl[1], P. Spégel[1], R. Lood[4], M. Shoeb[2], L. Chimuka[3], C. Turner[1]

Affiliations:[1]Lund University, Department of Chemistry, Center for analysis and synthesis, Lund, Sweden, [2]University of Dhaka, Department of chemistry, Dhaka, Bangladesh, [3]University of the Witwatersrand, School of Chemistry, Johannesburg, South Africa, [4]Lund University, Department of Clinical Sciences, Division of Infection Medicine, Lund, Sweden

According to World Health Organization, antibiotic resistance will be one of the main concerns in the near future. Little is known about how antibiotic resistance genes (ARGs) are horizontally transferred among bacteria. Bacterial stress is known to be related to ARG spread[1] however, to the best of our knowledge the driving force triggering bacterial stress has not yet been exhaustively studied.

The aim of this project is to determine the relationship between the chemical fingerprint in environmental water samples and bacterial stress. To achieve this, samples from three different countries collected with both spatial and temporal resolution will be examined using a multiplatform non-targeted analysis approach. Non-targeted analysis methods based on solid phase extraction - supercritical fluid chromatography – ion mobility – high resolution mass spectroscopy (SPE-SFC-IM-HRMS), which are currently being developed, will be applied in those samples to identify[2,3] small organic compounds. Additionally, different water parameters such as pH, conductivity or total dissolved solids are measured.

After gathering all these physicochemical information and measuring bacterial stress response, machine learning techniques will be applied to unravel correlations between the presence of pollutants and ARG spread.

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P55 - Chemical profiling of organic pollutants in the Vaal River Catchment in South Africa with possible links to antibiotic resistance using passive samplers and HPLC-MS

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The main threat to sustainable water supply in South Africa is the contamination of the water resources by pollution, of both organic and inorganic pollutants. The large levels of organic pollution in the environment and the consequent antibiotic resistance has become a huge public health problem globally. This work presents a detailed study which is based on both targeted and untargeted analysis of pharmaceuticals in one of South African rivers polluted by urban related activities. The selected study sites were along the Vaal River Catchment area located in Standerton, Mpumalanga; Villiers, Free State and Vanderbijlpark, Gauteng, which are provinces in South Africa. The target compounds were extracted from water samples using two analytical methods, i.e., passive sampling and solid phase-extraction (SPE) prior to their chromatographic determination by high-performance liquid chromatography coupled with tandem mass spectrometry (HPLC–MS/MS). Optimization of the SPE was achieved by spiking ultrapure water with 0.025 $\mu g\ mL^{-1}$ of the target compounds. Passive sampling (Chemcatcher® with a HLB receiving phase) was also optimized over a period of 14 days in order to obtain sampling rates (Rs) of the target compounds. The target compounds were detected in most of the river's sampling sites, using both solid phase extraction and passive sampling and will be discussed in the presentation.

P56 - Two-dimensional fractionation procedure followed by HPLC-HRMS as a tool for the identification of compounds from treated wastewater interacting with Aryl hydrocarbon receptor.

A, Vojs Staňová[1,2], Z. Toušová[3], M. Smutná[3], P. Mikušová[3], P. Šauer[1], H. Kocour Kroupová[1], K. Hilscherová[3] and R. Grabic[1]

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[2]Department of Analytical Chemistry, Faculty of Natural Sciences, Comenius University in Bratislava, Bratislava, Slovak Republic
[3]RECETOX, Faculty of Science, Masaryk University, Brno, Czech Republic

One of the most significant issues in analyzing environmental water samples is their complexity because they contain many substances of anthropogenic origin with different physicochemical properties. In addition, many of these chemicals present in complex samples are often unknown or arise because of various biotic and abiotic degradation/transformation processes of the original compound and may cause adverse effects to aquatic organisms at very low concentration levels e.g. especially endocrine disruption [1]. In this context, a combination of liquid chromatography and high-resolution mass spectrometry (LC-HRMS), allows the identification and quantification of many known and unknown compounds due to its high resolution, accuracy, and selectivity. From an analytical point of view, there are three approaches to LC-HRMS analysis of environmental samples, namely (i) targeted analysis, (ii) suspect screening, and (iii) non-targeted analysis [2]. The different fractionation procedures and bioassays are often combined in effect-directed analysis (EDA) to reduce sample complexity and identify the effect drivers.

In this work, we present an EDA study of treated wastewater extract eliciting Aryl hydrocarbon receptor (AhR)-mediated activity. AhR plays an important role in various physiological processes, including xenobiotic metabolism and immune response. Two-dimensional fractionation enabled separation of compounds in the total of four most active fractions, which were subjected to suspect and non-target screening using LC-HRMS. Advantages and limitations of the EDA approach coupled to non-target screening for identification of effect drivers in highly complex environmental samples are discussed together with an alternative approach to the separation of active compounds based on pull-down assay using specific ligand-protein interaction.

Acknowledgments

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P57 - Supercritical Fluid Chromatography in Hyphenation with Mass Spectrometry – New Gold Standard in Polarity-Extended Non-Target Screening

S. Bieber and T. Letzel,
both AFIN-TS GmbH, Augsburg, Germany

Non-target screening (NTS) is an analytical strategy, which can be used to detect and identify unexpected organic compounds, but also to compare small to large sample sets and to characterize them on a statistical basis. Depending on the question to be answered by NTS, the analytical process from sampling over analysis to data processing has to be adjusted thoroughly. Recent advances in the fields of chromatography, MS-instrumentation and data evaluation software have helped to improve the applicability of NTS as a screening technique.

However, the experimental analysis is still a challenging part in NTS if it comes to the analysis of (very) polar molecules. On the other hand, in the last decade the polarity-extended chromatography played an increasing role because non polar, polar and very polar compounds could be separated in one run. Specifically, RPLC-HILIC [1,2] and polar column supercritical fluid chromatography (SFC) [2,3] were very suitable strategies. Recent developments in special column combination [4], ionization prediction [5] and sensitive screening results [6] bring SFC now to a level of an excellent, sustainable and robust tool for non-target screening. The presentation will give a detailed overview in using SFC-ESI-MS as gold standard in various NTS disciplines like food and environmental analysis.

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P58 - ASSESSMENT AND EVALUATION OF MICROPLASTICS PARTICLES FROM GORAI BEACH OF ARABIAN SEA, MAHARASHTRA, INDIA: SEASONAL VARIATIONS AND CO-CONTAMINANT ESTIMATIONS

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Abstract

There is a growing concern worldwide about microplastics (MPs) and their effect on the environment. MPs have been detected in all the environmental matrices, ranging from aquatic, terrestrial, and atmospheric. However, these matrices are still not characterized thoroughly for the presence of different MPs. In the present study, the assessment and evaluation of MPs were done from the Gorai beach (19.2420° N latitude and 72.7808° E longitude), an Arabian sea coast in Mumbai, India. This is the first study on the evaluation of MPs from this beach in different season. The sample was collected during low tide, and a stretch of 600m was selected for microplastic sampling. A total of 2kg wet sand samples containing MPs were collected. The samples were dried in an oven at 65°C to reduce the moisture content. After drying, the samples were sieved with a mesh size of 5 mm to 0.03 mm. The visible MPs were then picked and segregated according to their morphotypes, size and colors. A total of 1756.6 particles/Kg dominated by polypropylene (PP) was observed in pre-monsoon. Maximum abundance of MPs have been observed during monsoon with the abundance of 7232.2 particles/Kg dominated by high density polyethylene (HDPE) and least in post-monsoon sample having abundance on 202MPs/Kg dominated by HDPE. The presence of heavy metals and organic contaminants were detected in different MPs particles. A total of 137 organic contaminants was extracted from MPs and Cr, Ni and Mn heavy metals were found to be present above permissible limit. This study showed that Gorai beach in the Arabian Sea is heavily contaminated with MP and persistent pollutants. There is an urgent need to remediate these MPs to protect the marine and surrounding environment.

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P59 - Uncertainty assessment of quantitative analysis of microplastics and Nano plastics using Py-GC/MS

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Abstract

Microplastics pollution is a recognized global threat that demands the development of new strategies to fulfill the growing need for standardized, rapid, and dependable measurements. Therefore, Accurate quantification of microplastics (MPs) and nano plastics (NPs) is essential to understand their distribution, environmental impact, and potential risks. This study focuses on assessing the uncertainty of quantitative analysis methods for MPs and NPs. Pyrolysis–gas chromatography–mass spectrometry (Py-GC/MS) is a technique with great potential for obtaining qualitative and quantitative data on MNPs mixtures through the selection of a set of characteristic pyrolysis products for each polymer. The uncertainty associated with the quantitative analysis of MPs and NPs with the Py-GC/MS is not well established. In this study we employ recovery experiments for the assessment of these uncertainties. We used a set of 8 internal standards at different concentrations and different matrices to comprehensively assess the impact of these parameters on the final results. Additionally, the LOD and LOQ values were estimated.

Keywords: Microplastics, Nano plastics, Py-GC/MS, Uncertainty.

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P60 - Micro Energy Dispersive X-Ray Fluorescence as a Tool for Monitoring the Migration of Brominated Flame Retardants Contained in Microplastics into Water Media: Case Study of Acrylonitrile Butadiene Styrene and Decabromodiphenyl Ether

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Microplastics (MPs) are not only considered dangerous for the environment due to their petrochemical origin, but also because they have the ability to release their manufacturing additives and concentrate pollutants from the surrounding environment. In addition, their small size makes them easily dispersed in all environmental compartments (water, soil and air), increasing the risk of ingestion by living beings in affected ecosystems, being accumulated and redistributed through food chains.

In this regard, brominated flame retardants (BFRs) have been the subject of study for many years due to their widespread use as additives in all types of plastics that are used in our daily lives (home furniture, upholstery, curtains, etc.). Studies have shown that these compounds cab cause significant toxicity in ecosystems where they are released, leading to restrictions or bands under current legislation. Specifically, one of the BFRs that has undergone legislative review is decabromodiphenyl ether (Deca-209), whose potential danger remained unnoticed for over a decade due to the challenges associated with its analytical detection.

Thus, this research focuses on studying the migration capacity of Deca-209, contained in acrylonitrile butadiene styrene (ABS) MPs, to aqueous media, and determine the effect of the most significant variables (salinity concentration and UV radiation) by means of a combination of Micro Energy Dispersive X-Ray Fluorescence (µ-ED-XRF) and Imaging Raman analysis.

The results obtained show the high potentiality of the µ-ED-XRF for the quantification of BFRs in MPs samples, and to monitor the dynamics of the release process, showing that low salinities (0.2%) and presence of UV radiation increase the transfer phenomenon. Lastly, the work here addressed evidence the advantages provided by Raman imaging to visually study the diffusion process of Deca-209 into the ABS matrix, showing the accumulation zones in the MPs crust that limit de migration.

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P61 - Automatic Raman Imaging Methodology for Microplastics Analysis in Complex Environmental Samples: Case Study of the Choqueyapu River basin (La Paz, Bolivia)

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Raman spectroscopy is a widely used non-destructive technique for identifying and quantifying microplastics (MPs). However, the MPs analysis from environmental water and sediment samples can often be challenging due to various factors. These mainly include the need to process a large number of samples, which can be time-consuming, the presence of background signals, fluorescence phenomena, and matrix interferences. Furthermore, variability in MP composition and the natural aging of the plastic matrix can further complicate the analysis, thus, potentially hindering its application and accuracy in the field.

This work aims to develop an analytical methodology based on Raman imaging analysis and chemometric models (Direct Classical Least Squares) to allow the automatic MPs characterization and quantification, and their classification by shape, colour and size. As a case study, the amount and type of MPs were determined in the Choqueyapu River basin (La Paz, Bolivia), being a strategic point of South America from which no data are still available. For this purpose, water and sediments samples from the river were collected, covering five significant points (n=3) along the basin. Subsequently, the sample treatment, filter support and measurement parameters (laser, spectral range, power, acquisition time and accumulations) were optimized to reduce the handicaps derived from these kind of environmental matrices, and to reduce the time of analysis to allow the fast and specific identification of the most frequently found plastics. Additionally, MPs databases (aged and non-aged) were prepared for the application of the classification models.

Overall, the results demonstrated the usefulness of Raman imaging as processing tool for fast MPs analysis if a controlled identifying method is specifically designed. In fact, the protocol here proposed was able to determine five types of plastics (polyethylene, polyethylene terephthalate, polystyrene, polypropylene and poly methyl methacrylate), their amount ($n^{o}_{particles}$/L or $n^{o}_{particles}$/Kg) and shapes (fibre, fragment and/or pellet). Thus, possible sources and causes of MPs in the area were also discussed at the light of the results obtained.

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P62 - Diagnosis of wastewater treatment plants (WWTPs) efficiency for the treatment of microplastics (MP) in Wallonia, Belgium: methods and preliminary results

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The occurrence of microplastics (MP) in the environment has become a global environmental problem. A rough estimate predicts that 70-80% of marine plastics originate from land and are carried by rivers to the oceans (Wagner and al., 2014). Currently, there is a rising concern on the role of wastewater treatment plants (WWTPs) in the release of MP in the environment. Recent studies have shown that the presence of MP in wastewater is partly due to plastic microbeads contained in personal care products, but also to plastic fibres released by washing synthetic fabrics (Brown DM and al., 2001). During treatment process at WWTP, around 99% of MP move mainly to sludge (Lares M and al., 2018).

The MicroPlaSTEP project aims to assess the efficiency of Walloon WWTPs for the treatment of MP. First, a protocol of samples preparation has been developed to remove organic matter and separate MP by density difference. In a second time, a representative selection of WWTPs in Wallonia has been characterized in terms of MP load in wastewater (inlet), in effluent (after treatment) and in sludge. Two analytical techniques have been used to evaluate the number of MP according to their size: (1) Nil Red Microscopy for particles from 100 µm to 1 mm, and (2) FTIR microspectrometry from 20 µm to 100 µm. In addition, FTIR microspectrometry gives the type of MP (PE, PP, PS…). The poster will present the protocol of samples preparation and first results of MP distribution in the Walloon WWTPs MP characterisation by these two techniques.

Microscopy with Nil Red

Microspectrometry FTIR

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P63 - First Assessment of Metal Presence on Microplastics on the Cantabrian Coast (Northern Spain): Analytical Methodology Based on Hyperspectral Imaging and Inductively Coupled Plasma-Mass Spectrometry

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Several researches show that microplastics (MPs) present a risk as a reservoir of contaminants, being able not only to release their manufacturing additives along the environmental compartments, but also to adsorb the pollutants present. Thus, they are a route capable of increasing the distribution, bioaccumulation and ecotoxicity of pollutants. However, their potential as environmental indicators by their container role of metals, metalloids, and/or technological metals have not been yet exploited, as most of the studies are often focused on MPs monitoring to identify and quantify which MPs are present instead of what they carry.

This study aims to answer to the following, which and how many MPs are arriving to our coast? Which is metallic level that our beaches are receiving by what is adsorbed on MPs?

For this purpose, a bi-annual sampling campaign was performed in three significant points of the Cantabrian Coast to cover pristine, touristic and urban scenarios (Sonabia, Sopelana and Areeta, respectively). As the MPs role as reservoirs depends on their surroundings but also on the nature of the plastic matrix, in a first step, a non-destructive characterization method based on Hyperspectral Imaging (HSI) was used to quantify and classify the MPs collected. Subsequently, an acidic metal leaching method was optimized to minimize the time-consumed per analysis, and the leachates were analysed by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). Finally, chemometric tools were used to deeply study the results by Principle Component Analysis (PCA).

This study overall shows the usefulness of (HSI) for a fast MPs identification of the most common MPs found on beaches (polyethylene, polypropylene and polystyrene), and allows to evaluate the risk associated with the presence of metals adsorbed on MPs, specifically in a highly populated area with important tourist activities and former industrial activities.

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P64 - Quantitative NMR of PET micro particles in water for the characterization of a candidate reference material.

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Because micro plastics in the environment move more and more into the focus of the regulatory bodies, there is a strong need for reliable, harmonised measurements. Therefore, the Joint Research Centre of the European Commission in Geel has set out to produce certified reference materials (CRM) of micro plastics for that purpose. One such candidate reference material is polyethylene terephthalate (PET) in water to support EU policy.

For the characterisation of this material a method employing quantitative nuclear magnetic resonance spectroscopy (qNMR) with internal standard reference material (ISRM) was developed. When the ISRM is a CRM itself then this mode of qNMR is a potential primary ratio method with shortest possible connection to the International System of Units (SI). The method builds upon what Peez et. al.[1] reported. In brief, the PET containing water is filtered to collect the particles on a filter disk. The disk with the particles is transferred into a glass beaker and a precisely weighed amount of ISRM is added. PET and ISRM are dissolved off the filter disk with d_1-chloroform/ trifluoroacetic acid (4/1, v/v). This solution is then measured with 1H-NMR.

The poster will display details of the method, the choice of ISRM, why not all requirements for primary ratio method are met, pitfalls during development, and a budget of the uncertainty contributions

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P65 - Microplastic Characterization and Screening by Combining DART and High-Resolution Mass Spectrometry

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Introduction

Microplastics (small plastic particles ≤ 5mm) result from commercial product development and the breakdown of plastics and are abundant in our environment from variety of sources. Commercial sources include cosmetics, textiles to other pieces of plastics such as water bottles that breakdown with radiation. Microplastics have been detected at an alarming level in our marine life and drinking water. Direct Analysis in Real Time (DART)-high resolution mass spectrometry allows for a rapid fingerprinting of environmental microplastics and the screening of additives. Typical samples of interest are polymers found in the environment like virgin pre-production pellets, microbeads from personal care products, microplastics found in the aquatic environment, and synthetic fibers.

Objective

Fingerprinting for the identification of the origin of microplastics by comprehensive screening with thermal desorption coupled to fast and direct DART-HRMS analysis.

Methods

DART was coupled to an impact II VIP QTOF (both Bruker, Germany). While traditional GC/MS methods require sample preparation (10-20 min) and long analysis times (> 20 min), this new method allows for rapid polymer analysis (< 5 min) without sample preparation. A small sample sliver is cut and placed in a copper pot of the IonRocket thermal desorption system (Biochromato Inc., Japan) which delivers temperature gradients from ambient to 600 ºC with ramp rates of 150 ºC/min. When the thermal program is started, the QTOF collects MS spectra at 5Hz for the entire run.

Results

The resulting mass spectra display thousands of discrete peaks. Depending on the temperature, different additives, the polymeric basis as well as degradation products are released and detected at different time points during the run. Distinct m/z signals and general signal patterns of a microplastic material found in water were compared with standard plastic samples such as a soda bottle, a trash bag and packaging material.

Discussion

While the soda bottle turned out to be made of PET, the microplastic clearly had PE as a basis. PE as an insoluble polymer is difficult to analyze with other MS-based methods like MALDI. The microplastic material showed most similarity with the trash bag, with regard to both certain distinct marker m/z values as well as the general pattern released by the temperature gradient, so it can very likely be traced back to that origin. The combination of DART with HRMS allows for rapid fingerprinting of environmental microplastics and the identification of unknown compounds with high resolution and high mass accuracy.

P66 - Quantifying Microplastics abundance and their implications as vectors for heavy metal adsorption in freshwater aquatic system of Kashmir Himalaya

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Abstract

Microplastic (MPs) contamination in the freshwater systems has recently attracted widespread attention but, the distribution, morphology and interaction of MPs with heavy metals on freshwater aquatic systems has remained elusive and poorly understood. The present study examines the MPs abundance in sediment and water samples and factor controlling their distribution in Manasbal Lake, north-western (NW) Himalayas. The MPs abundance ranged from 13 $n \cdot L^{-1}$ to 89 $n \cdot L^{-1}$ (average=46.88 particle/L) in lake surface water samples, while their concentration varied from 840 $n \cdot kg^{-1}$ to 4020 $n \cdot kg^{-1}$ in sediment samples, with an average abundance of 2177.6 particle/kg in the dry sediments. Five main types: beads/pallets/granules, fragments, fibres, foams and films were found, wherein beads dominate the quantity accounting 92.24 % of the total microplastics in the sediments. Polypropylene, polyethylene and polystyrene were observed dominant polymers suggesting the pathway through domestic sewage effluents, tourist and fishing activities. No significant correlation was observed between microplastic abundance in surface water and sediment samples. The MPs abundance in Mansabal Lake were closely related to distance from the lake inlet. Furthermore, we also looked into the role of MP morphology in heavy metal adsorption. Scanning electron microscopy with energy dispersive X-ray spectroscopy (SEM-EDS) results indicated that the presence of eleven heavy metals (Cu, Cd, Zn, Ni, Co, Fe, Mn, Cr, Bi, Hg and Pb) on the surfaces of different MPs in the lake. We conclude that MPs in sediments act as carriers of heavy metals and activate their bioavailability, hence posing a potential threat to the ecological security of water body.

Keywords: Freshwater lakes; Grain size; Himalaya; Microplastics; Total organic carbon

P67 - New sampling technique for determination and identification of microplastics on plants with chemical imaging by FTIR- and Raman microscopy

Yasmin Oguz, Mary Yeboah Obiri, Justus Schöttl, Mike Wenzel, Gerrit Renner, Thorsten Schmidt, Jürgen Schram

Microplastics in our environment are contaminants increasingly caused by our throwaway society. They can come into agricultural products through different processes, like the deposition of airborne microplastics, plowing under foils into the soil, or microplastics from aquatic systems used for watering. In most of these cases, microplastics accumulate on the surface of agricultural products.

A newly developed exfoliation technique with well-defined glass beads (µpeel) separates microplastics and other particles from the plant surface into a glass bead phase. This solid phase afterward is transferred into a recently developed microplastic separator for sediments (and glass beads), using flotation with air bubbles to separate even microplastics with heavy density from the aqueous solution or suspension. It enriches it quantitatively on a metal sieve plate (*µsep*) ready for further analysis [1,2].

With an FTIR microscope and Raman microscope, analytical imaging of the filter surface is processed using a recently in our group developed robust, fast, and open-source database microplastics identification software programmed with Python (µident) [3,4]. In addition, a further open-source Python-based tool (µmap) controls the image scanning of the FTIR-microscope, automatically recognizing the spot to be analyzed intensively and thus accelerating the capture of an image of the microplastics distribution on the surface of the sieve [5].

The deposition of microplastics on the surface of different (food)plants could be determined, thus revealing the possibilities of this new integral method to determine microplastics on plants.

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P68 - Nanoplastics analysis in bottled water: insights from Atomic Force Microscopy Infrared Nanospectroscopy

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In recent years, microplastics (MPs) and nanoplastics (NPs) have emerged as ubiquitous contaminants hinting to a high likelihood of human exposure. NPs have attracted the attention of the scientific community since their sub-micrometre size increases their potential to cross biological membranes, and their translocation and accumulation in human tissues can lead to toxic effects on human body.

The physical and chemical characterization of NPs is key for a better understanding of their exposure, fate, and potential implications for human health. Bulk and micro-scale analytical methods provide rich information about bulk synthetic polymer samples and even single MPs, but lack the spatial resolution required to gather insights into presence of NP contamination.

In our work, we demonstrate the capabilities of infrared nanospectroscopy for the nanoscale physico-chemical characterization of NPs. We will showcase the enhanced spectroscopic sensitivity and single-nanoparticle spatial resolution provided by the unique combination of atomic force microscopy and infrared spectroscopy, also in comparison with a range of state of the art micro-analytical techniques. We demonstrate how this method enables a thorough multidimensional study of different synthetic polymers in the form of NPs. We then illustrate the capabilities of this strategy by analysing NP contamination in a set of bottled water samples.

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P69 - Non-Targeted Detection of Virgin Olive Oil adulteration using 1D NMR and FTIR Spectroscopy combined with Chemometrics

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The significance of detecting adulteration in olive oil has lately risen because producers are becoming more and more attracted to this illicit practice. As a result, improved analytical techniques for spotting olive oil adulteration are now required. In this study, FTIR and one-dimensional (1D) nuclear magnetic resonance (NMR) techniques combined with multivariate statistical analysis are used to demonstrate an efficient method for detecting adulteration in olive oil that has been intentionally tampered with by adding varying amounts of other edible oil. A total of 150 samples of adulterated virgin and organic extra virgin olive oil were examined. Virgin and organic extra virgin olive oils were spiked with various edible oils at quantities ranging from 0% to 75% v/v. In comparison to other approaches that have already been published, these methods are straightforward sample preparation, quick sample analysis, new process parameters, and simple-to-understand results, which make it an accessible tool for detecting olive oil fraud by substitution or dilution with other edible oil.

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P70 - Rapid discrimination of black truffles and annotation of unknown marker compounds by DART-QTOF-MS

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Food fraud is a major issue in the food industry leading to financial losses for food processors as well as inflicting lasting damage in the trust of consumers. Due to the motivation of financial profit, particularly expensive products are prone to adulteration. Truffles are considered a luxury product with prices ranging up to 1000 - 2000 €/kg for the Périgord truffle (*Tuber melanosporum* VITTAD.). Other black truffle species, for example the Asian truffle (*T. indicum* COOKE ET MASSEE) are morphologically highly similar to the Périgord truffle but much cheaper in price. Therefore, identification of adulterated truffle batches is of utmost importance, leading to a demand for analytical solutions that deliver highly reliable results, while simultaneously being easy-to-operate and providing a high degree of automation for high-throughput analyses.

In this study, we developed a comprehensive workflow for the differentiation of different black truffle species utilizing direct analysis in real time (DART) ionization (DART JumpShot source, Bruker Daltonics) in combination with QTOF-MS (Impact II, Bruker Daltonics). Unsupervised and supervised multivariate statistical models built using the software MetaboScape 2023b (Bruker Daltonics) revealed a successful discrimination of the truffle samples according to their species. In the next step, the marker compounds with the largest contribution to the species discrimination were annotated using tools for untargeted unknown identification included in MetaboScape. In detail, annotation was performed based on the information of accurate mass, isotope pattern, and fragmentation pattern, either by a fully automated spectral library search or by a semi-automated annotation workflow comprising elemental composition prediction, structure assignment and *in silico* fragmentation.

Compared to chromatographic methods, DART-QTOF offers significantly shorter analysis times of 15 s per sample and reduced solvent consumption. All in all, DART-QTOF paired with chemometrics presents a fast, robust, and resource-saving method to counteract adulteration of black truffles.

P71 - Chemometrics to differentiate hop cultivars through volatile fingerprint

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Hop is traditionally used in beer mainly due to the content of bitter acids, but the increasing consumer interest and higher production in craft beers, essential oils covered relevance since they are secondary metabolites of the hop plant responsible for beer aroma and flavor [1]. In this study, an untargeted chemical fingerprint with a subsequent chemometric analysis was used to evaluate the differences in volatile organic compounds (VOCs) among four different hop cultivars from two growing areas in the Paraná state of Brazil.

Hop cones from different cultivars were analyzed using purge & tramp coupled to gas chromatography-mass spectrometry (P&T-GC/MS). VOCs were extracted and concentrated from 200 mg of each sample warmed until 60ºC for 30 min of purge before the desorption for the GC-MS analysis. The identification of VOCs was accomplished based on similarity and fragmentation patterns with mass-spectral libraries and the retention indices. To manage the data obtained it was used the collaborative online resource Workflow4Metabolomics (W4M). The webpage Flavornet was used to assess the aroma descriptors of each VOC.

The results have shown a wide range of VOCs in all hop cultivars, mainly terpenes. Humulene, ß-myrcene and caryophyllene were some of the most concentrated volatile odorants, providing a woody and spicy aroma. The use of chemometrics enabled to determine the differences in the number of volatile compounds obtained among the cultivars, as well as the intensity of the compounds in common. These analyses are an approach for further markers development that might explain the different chemical compositions among hop cultivars.

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Acknowledgements:

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P72 - NANOCRYSTALLINE GRAPHENE SENSOR FOR EXTRA-VIRGIN OLIVE OILS TESTING

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Olive oil is one of the most valuable foods due to its unique organoleptic properties, its high content of monounsaturated fatty acids (70-80%) and its antioxidant properties given by the presence of lipophilic phenols (tocopherols) and hydrophilic phenols (hydroxytyrosol, tyrosol, oleacein, vanillic acid) in the composition of olive oil. Extra virgin olive oil (EVOO) is traded at higher prices than other vegetable oils due to its exceptional properties such as nutritional value, aroma and excellent taste and is therefore subject to fraudulent activities. Therefore, we aim to develop an electrochemical sensor targeting the adulteration of extra virgin olive oil. The proposed analytical approach will be used to determine the authenticity of extra virgin olive oil. The active material used as transducer is based on nano-crystalline graphene (NCG) to obtain sensitive and conductive surfaces for the detection of organic compounds in olive oil, including extra virgin olive oil, to determine the authenticity of the oils of extra virgin olives. The growth of these nanocrystalline graphene films is achieved by the plasma enhanced chemical vapour deposition (PECVD) process. The growth process is influenced by the conditions under which the dissociation of the starting materials used (in this case it is methane - CH_4) takes place in the plasma phase. An important factor is also the adsorption of carbon-based radicals on the surface of the substrate, where they generate nucleation centres and form two-dimensional structures of rings consisting of sp2-hybridised carbon atoms, due to the surface charged with energy and bombardment with ions from the plasma. After the appearance of these nuclei on the surface of the substrate, the process continues with the formation of edges due to the lateral growth of the nuclei, creating a continuous film until the substrate is completely covered.

The characterisation of the developed sensors (N-NCG-2/N-NCG-3) to be used for the determination of the authenticity of extra virgin olive oils was done by comparing the voltammogram of 5 mM potassium ferricyanide with the voltammogram obtained for the classical glassy carbon electrode. The following figure shows the sensitivity of the developed sensors compared to the classical glassy carbon electrode. The developed electrochemical sensors were applied to determine the authenticity of extra-virgin olive oils.

Acknowledgments: This work was supported by a grant of the Ministry of Research, Innovation and Digitization, CCDI – UEFISCDI, project number PN-III-P2-2.1-PED-2021-2256, within PNCDI III and by the Core Program within the National Research, Development and Innovation Plan 2022-2027, carried out with the support of MRID, project no. 23020101(SIA-PRO), contract no 7N/2022.

P73 - A multi-step sample preparation coupled with gas chromatography-ion trap mass spectrometry for the identification and quantification of rotundone in grapes

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The aromatic component is a fundamental parameter in the consumer's appreciation of wine since first impressions are often dictated by olfactory examination. Wine is one of the most complex agri-food products as it contains almost 900 compounds, of which only 10% are likely to contribute to the aroma. Among them, rotundone was first identified in 2008 as the aromatic compound responsible for peppery notes in red wines. It is often perceived positively by connoisseurs consumers. Rotundone is an extremely potent aroma, with olfactory detection thresholds of 8 ng/L in water and 16 ng/L in red wine.

This work describes the development and validation of an analytical method for the quantitative determination of rotundone in grapes. The method uses stable isotope dilution analysis with *d5*-rotundone as internal standard, solvent extraction followed by solid-phase extraction and then direct immersion-solid phase microextraction. The analysis is performed by gas chromatography–ion trap mass spectrometry. Various data acquisition methods were studied (SCAN, SIM, MS/MS). The tandem mass study provided new insights into the molecule's fragmentation mechanisms. The analytical method was validated on the basis of linearity, limit of quantification, fidelity, accuracy. It was then applied to grapes of different varieties in order to evaluate the varietal capacity to produce rotundone. The concentration range obtained varies from <LOQ (10-30 ng/kg) to 700 ng/kg.

P74 - Bio-based and biodegradable polymers for food contact: a chemical approach to investigate their safety

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Bio-based polymers derived from renewable biological resources and biodegradable polymers that will easily disintegrate and biodegrade in the environment, are being developed and promoted as an alternative to conventional petroleum-based non-biodegradable plastics to be utilized in food packaging. Some of the most common bio-based and biodegradable polymers are those based on PLA, PHA, PBS, and so on. However, these materials generally do not perform as well as conventional plastics and require additional chemicals such as plasticizers, antioxidants, light and UV stabilizers, release agents, crosslinking agents, etc. Alternatively, the polymers are blended together or copolymerized to obtain materials with improved properties. Therefore, these polymers, like other food contact materials, can release low molecular weight components to food and pose a health risk to consumers. The chemical safety of these sustainable materials has been scarcely studied.

In the framework of MIGRABIOQUANT project, funded by the Spanish National Research Agency, an analytical methodology to identify potential migrants in bio-based and/or biodegradable materials is being developed. The approach involves the combination of several techniques, namely FTIR for polymer identification, GC-MS for volatile and semi-volatile migrants and LC-MS for non-volatile migrants. In a small sampling carried out, polyester based materials and PLA, were some of the most common marketed materials. Regarding the potential migrants, it is worth noting the identification of NIAS, thus, for example, different oligomers in the samples analyzed. However, more research is required since data on migration in foods and human exposure assessment is scarce and they are essential elements for a complete risk assessment.

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P75 - Risk assessment of plastic breastmilk bags. Evaluation of non volatile migrants.

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Food safety is crucial to people's well-being. That is why, when storing food in a plastic container, it is necessary to ensure that no compounds are transferred that could harm the consumer's health. Therefore, plastic materials intended for contact with food must comply with Commission Regulation (EU) No 10/2011[1].

In this study, eight commercial bags intended for the storage of breast milk were analyzed. The non-volatile compounds present in these bags and their migration into the food simulant assigned for milk in the regulation (50% ethanol) were identified and quantified. The analyses were performed using ultra-high performance liquid chromatography coupled with mass spectrometry (UPLC-MS), working with two types of mass analyzers: a triple quadrupole (QqQ) and a quadrupole time-of-flight (QTOF-MS).

The results showed the migration of known compounds such as fluorene (a precursor in the production of inks and adhesives) and some common slip agents in polyolefins like oleamide and erucamide. Several non intentionally added substances (NIAS) were also detected, such as some degradation productos of plastic aditives. Specific toxicity studies should be carried out to ensure that there is no toxicological risk to infants.

[1] Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food.

P76 - Heterocyclic aromatic amines in heat-treated processed cheese

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Text: Regular intake of dairy food is desirable due to the content of many essential nutrients (Comerford et al., 2021). For the preservation of dairy foodstuffs, heat treatment is often used which, however, can lead to the production of potentially harmful substances including heterocyclic aromatic amines (HAAs) (Chen et al., 2020). There is only limited information available on the content of HAAs in preserved dairy products. Here, thermic HAAs amino-imidazo-azaarenes were monitored in heat-treated canned processed cheese using LC-MS. Preliminary results indicate the presence of IFP (2-amino-1,6-dimethyl-furo[3,2-e]imidazo[4,5-b]pyridine) in heat-treated processed cheese, in contrast to untreated processed cheese (Fig. 1).

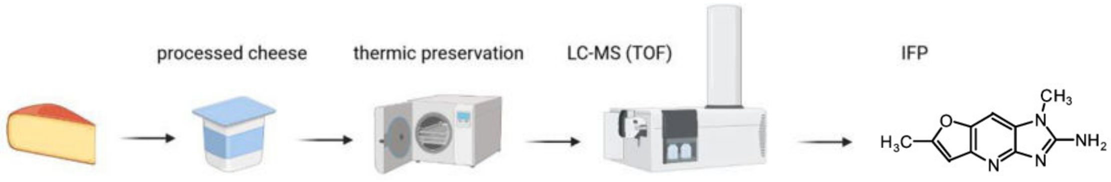

Figure 1. Schematic representation of the experiment design. Created in BioRender.com.

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P77 - Identification of non-volatile compounds in bio-based and/or biodegradable packaging materials using a non-targeted LC-HRMS method

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Through the “Farm to Fork Strategy” the EU promotes the use of sustainable packaging solutions utilizing environmentally friendly materials. Bio-based and biodegradable polymers are being developed and already being used to replace petroleum-based plastics for food packaging applications, it is expected to continue to grow. However, it is important to highlight that the critical issue of chemical safety is often overlooked. The migration of compounds from the packaging can affect food safety and needs to be evaluated. The potential migrants include IAS such as monomers and additives and the so-called NIAS, such as reaction and degradation products and impurities. In the present work, a non-targeted analysis using LC-HRMS to investigate non-volatile compounds extracted from packaging materials labeled as bio-based and/or biodegradable and intended to be in contact with food was performed. The samples, characterized by FTIR, include polyester based materials, polylactic acid (PLA), and polypropylene. After extraction with acetonitrile at 40 ºC for 24 h, the resulting extracts were analyzed by Q-Exactive-LCMS. Data were acquired in full scan mode and processed using an interface under the open-source programming R environment [1]. Both IAS, such as the additives antioxidants (e.g., Irganox 1076), slip agents (e.g., erucamide) and plasticizers (e.g., DEHP), and NIAS were detected in the samples. Among NIAS, oligoesters based on different combinations of diols (ethylene glycol 1,3-butanediol, neopentyl glycol, etc.) and diacids (adipic, phthalic, and sebacic acids) were tentatively identified. As in a previous study, most of them presented a cyclic structure [2]. Particular attention should be paid to NIAS, since most of them are unknown compounds and their toxicity has not been evaluated.

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P78 - Non-targeted screening for volatile compounds in food contact bioplastics using Purge and Trap coupled to Gas chromatography-Mass Spectrometry

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Bio-based and/or biodegradable plastics are being developed as sustainable packaging solutions. The use of environmentally friendly bioplastics is expanding, and it is expected to grow significantly in the next few years. Similarly to petrochemically based polymers bioplastics are subject to the regulatory requirements of food contact materials in terms of inertness and safety of the material. Both intentionally (IAS) and non-intentionally added substances (NIAS) should be evaluated to protect the consumers' health. Non-targeted methods are commonly used for the analysis of NIAS. Sample preparation is the first step in the analysis of potential migrants and usually it is a laborious and time-consuming procedure. The Purge & Trap system coupled to Gas chromatography-Mass Spectrometry (P&T-GC-MS) has demonstrated to be an efficient analytical tool for the extraction and subsequent analysis of volatile compounds.

In this work, a P&T-GC-MS method was optimized for the analysis of volatile compounds in food packaging samples labeled as bio-based and/or biodegradable and based on polypropylene (PP), polyester and Polylactide (PLA). Samples were subjected to 80 ºC for 30 min and the separation of the analytes was made using a Rxi-624Sil MS (30 m × 0.25 mm internal diameter, 1.40 µm film thickness) column. The NIST/EPA/NIH 11 (version 2.0) and Wiley Registry^{TM} 8th edition mass spectral libraries were used for identification purposes. Only 16 of the 120 compounds detected in the samples are listed in the Regulation 10/2011. Monomers such as propylene glycol and 2-ethyl-1-hexanol, were identified in polyester samples. 2,3-pentanedione was identified in all PLA samples and it has been described as a degradation product [1].

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P79 - Solid Phase Extraction (SPE) method for analysis of BISPHENOL A (BPA) and Bisphenol A diglycidyl Ether (BADGE) in various food matrices based on Molecularly Imprinted Polymers.

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Bisphenol A is an endocrine disruptor which can mimic the body's own hormones and may lead to negative health effects. The migration of BPA from the packaging to food is the main source of consumers' exposure to BPA. BADGE is also potential migrant into the preserved food and is of toxicological concern. Bisphenol A (BPA) and Bisphenol A Diglycidyl Ether (BADGE) are molecules widely used in food contact materials.

In April 2023, EFSA published a reassessment of the safety of BPA, significantly reducing the Tolerable Daily Intake (TDI) set in its previous assessment in 2015 from a limit of 4 µg per Kg of bodyweight per day to 0.2 ng per Kg of bodyweight per day. The TDI is therefore around 20,000 times lower than the previous one.

In this work, molecularly imprinted polymers were used for the solid phase extraction (SPE) and concentration of bisphenol A and BADGE from various food matrices: water, milk, cola drinks, beers: infant formula (liquid and solid), canned food. Thanks to selectivity of MIP, a strong clean-up and high recoveries (>80%) were obtained. The analyses were carried out by LC-Fluorescence.

P80 - First approach on the stability and bioaccessibility of epoxy-resin contaminants from packaging materials in food

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Epoxy-based coatings applied to the inner surfaces of cans into contact with foods represent an important source of contamination through the migration of chemicals into food. The present study was focused on the main compound in epoxy resins, Bisphenol A DiGlycidyl Ether (BADGE), which results from the condensation of epichlorohydrin and bisphenol A (BPA) [1].

The aim of this work was to evaluate the in vitro bioaccessibility of BADGE, as well as to study its chemical stability during the gastrointestinal digestion process following the standardized INFOGEST protocol [2]. A suitable chromatographic method was developed to identify the compounds of interest and evaluate the effect of the food matrix components, such as the presence of amino acids, as well as the activity of enzymes on BADGE bioaccessibility. The results showed that 60% of BADGE ingested is bioaccessible for its absorption after digestion. The presence of amino acids does not seem to significantly affect the final bioaccessibility once in the digestive tract (around 50%), except in the case of histidine (18%). The formation of small amounts of products derived from BADGE with chlorohydrins (BADGE·HCL and BADGE·2HCl) was observed at the gastric phase, though its bioaccessibility was reduced during the intestinal phase. Further research should be carried out to study different pH conditions and evaluate the effect of other food matrices on the BADGE-related compounds formation during gastrointestinal digestion with potential toxicity.

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Acknowledgement:

BACFood4Expo project (PID2020-114569RJ-I00) funded by Spanish Ministry of Science and Innovation under the competitive State R&D Program Oriented to the Challenges of the Society, “R&D Projects 2020” - Modalities “Research Challenges and Knowledge Generation”.

P81 - Validation of the analytical method for inpyrfluxam in agricultural products using QuEChERS and LC-MS/MS

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Inpyrfluxam is a pyrazolecarboxamide fungicide discovered by Sumitomo Chemical in 2020 and belongs to the succinate dehydrogenase inhibitor (SOHi) group of fungicides. It is effective to protect against Rhizoctonia species, which cause seed decay, seedling damping-off and root rot. In this study, a QuECheERS(EN method and PSA with MgS04 clean-up) and LC/MS/MS for the determination of inpyrfluxam in agricultural commodities was validated according to Codex guidelines(CAC/GL 40). The five representative food commodities were used to verify the present method, which obtained good linearity(R^2>0.99) in the 0.001-0.075 mg/L calibration. The mean recovery for inpyrfluxam at three concentrations(LOQ, 1QxLOQ, 50xLOQ, n=3) was in the range from 84.1 to 113.2% with the relative standard deviations less than 6.8%. And the limit of detection and limit of quantification for inpyrfluxam were below 0.003 and 0.01 mg/kg, respectively. Based on these results, this proposed analytical method found to be appropriate for inpyrfluxam determination and can be served as a reference method for the establishment of maximum residue limits(MRLs).

P82 - Losses of sulphur volatile compounds from freshly roasted coffee ground at different temperature

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Text:

Flavour is once from the most important aspect of a good coffee for the consumer. Volatile compounds are the main ingredients in creation coffee flavour and aroma. Their decrease during coffee grinding should negatively affect the overall enjoyment of coffee. Freshly roasted high-quality arabica coffee beans were ground at in cryo mill (-196 °C) and also at ambient temperature (22 °C) in tube mill. Samples were taken at the start and then every 4 hours, vials open to air. All samples were measured on GC-MS and GC-FID-SCD in order to track the loss of odorous sulphur compounds of varied volatility such as methanethiol, dimethyl sulphide, dimethyl disulphide, thiophene and 2-methyl thiophene.

GC-MS results show that for coffees ground at cryo temperature, zero-time peak areas of *all* volatile compounds were in average *1.8-times* higher than the ones treated at ambient temperature. Consequently, the peak intensity declined much faster fast, namely, in first hour, in the cryo-treated samples and after 4 hours the concentrations of all sulphur compounds were similar regardless the grinding temperature. 99 out of total 109 compounds were identified and sorted in which pyrazines dominated making up the typical aroma of roasted coffee beans, followed by ketones bringing more buttery, creamy and caramel notes. Sulphur compounds were in minority (<1 %) yet yielding essential coffee aroma. Taking advantage of a selective SCD, the marginally abundant sulphur-components could be detected and identified despite their co-elution with other non-sulphur components with less odour impact (hits by SCD/MS ~ 17/8). Cryo-milling offers a mild method preserving the sulphur volatiles. Decline in the intensity of volatiles was well observable on GC-MS as well as 2D GC system. Additionally, SCD detector is perfect for monitoring of sulphur compounds, as it captures all sulphur compounds (potentially hidden on 1D GC-MS system). All in all, the methods are complementary and both are needed for reliable identification.

P83 - Analysis of pesticide residues in Fruit and Vegetables using LC HRMS

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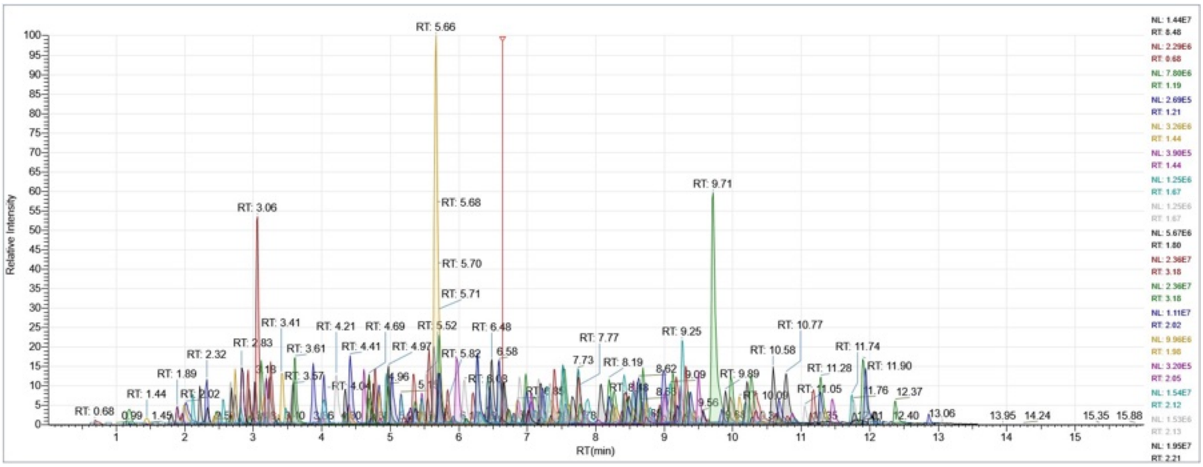
Text: The Fruit and vegetables are extracted using Dutch Mini Luke method; after extraction with acetone/dichloromethane/petroleum ether 40-60°C a solvent exchange into ethyl acetate is carried out. The ethyl acetate fraction is analysed by GCMS. A portion of this fraction is diluted 1 in 10 with methanol for LC HRMS analysis.

The DAFM laboratory is currently transitioning the Fruit and Vegetable analysis from QQQ MS to HRMS. The LC HRMS method was accredited for 268 pesticides in 2021. Last year the scope was expanded by 40 pesticides. The validation was carried out according to SANTE/11312/2021. The following parameters were assessed, Linearity, Repeatability, Reproducibility, Mass accuracy, Reporting levels and Matrix effects.

The method consists of 16 min LC gradient and positive full scan with vDIA experiment. HRMS allows for easier addition to the scope as no tuning is necessary, only target and confirming ions selection is required. Full scan acquisition allows for potentially unlimited scope expansion as well as retrospective analysis. The untargeted screening capabilities of the system provide the possibility of identifying emerging pesticides that are not on the laboratory's scope.



References: Extraction method is based on standard multi-residue methods published in "Analytical Methods for Pesticide Analysis 6thEd" – Ministry of Public Health, Welfare and Sport, The Netherlands.

P84 - Determination of benzimidazoles and ciprofloxacin in milk by dispersive microextraction with electrochemical detection based on carbon nanotubes modified with electropolymerized L-DOPA

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The presence of active principles for veterinary use in food is a reality that leads to the generation of public policies tending to define the maximum residue limits. In this scenario, it is extremely important to have simple detection and quantification methods for multi-analyte analysis. Within food matrices, due to its components, milk is a highly complex sample, so the pretreatment steps for subsequent detection with complex methods, such as chromatography, are complicated. In the present work, a dispersive microextraction method was developed for the extraction of albendazole (ABZ), febendazole (FBZ), thiabendazole (TBZ) and ciprofloxacin (CIP), making use of the effervescent capacity of sodium carbonate to achieve the best dispersion and extraction of the analytes of interest in the extracting solvent. In parallel, a sensor platform based on carbon nanotubes modified with electropolymerized L-β-3,4-Dihydroxy-phenylalanine (L-DOPA) was developed to generate a melanic polymer. In general, conjugated compounds having polyaromatic rings and functional groups bind to the sp2 lattice of graphene sheets of the nanotube by π-π stacking, and the functional moieties stabilize the sp2 sheets in the solvent and provide new functionalities to the nanomaterial[1].Therefore, the study of the interactions between melanic polymers with different substrates has been implemented over the years for various purposes, including surface functionalization[2]. Finally, the combination between the microextraction system and the electrochemical detection allowed, thanks to the preconcentration factor, to obtain good detection limits. The method was validated obtaining good results. The complete system was tested with samples from different sources, in order to determine the presence of the compounds in commercial milk samples.

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P85 - Monitoring of veterinary drugs in livestock products using a simultaneous multi-component analysis method(LC-MS/MS)

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Veterinary drugs are used for the prevention and treatment of diseases that occur in livestock farming. Excessive use of animal veterinary drugs can lead to residue issues, increased antibiotic resistance, and potential impacts on human health. In Korea, the Positive List System (PLS) will be implemented starting from 2024. This system aims to prevent misuse of animal veterinary drugs and confirm the use of unregistered or prohibited drugs. To enable rapid testing, Simultaneous multi-component Analysis Method(LC-MS/MS) was developed targeting five representative livestock products(beef, pork, chicken, egg, milk). This method allows for the simultaneous analysis of 156 kinds of substances available livestock. Through the CODEX guidelines (CAC/GL 71-2009), the developed method was validated to be able to detect and quantify veterinary drugs with good linearity, precision, and accuracy. R-square value (coefficient of determination with calibration range) of the linear
standard calibration curves was higher than 0.99. The recovery rates were ranged from 60% to 120%, with the coefficient of variability(CV) of less than 32%.

Monitoring was conducted on 100 samples of domestically distributed livestock products in korea. The results showed that the residues were below the Maximum Residual Limit(MRL), indicating that they were being safely managed.

P86 - Optimization of deep eutectic solvent-based ultrasound-assisted emulsification microextraction for the rapid determination of benzophenone-type ultraviolet filters in tea beverages

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Benzophenone-type ultraviolet filters (BP-UVFs) are a group of emerging contaminants with health concerns, which found in tea beverages raising potential risks for food safety and could bioaccumulate in the food chain. This study describes a simple method using a “green” deep eutectic solvent (DES) based-ultrasound-assisted emulsification microextraction (USAEME) to rapidly extract BP-UVFs from tea beverages, and they were then detected and quantified by UHPLC-electrospray ionization (+)-quadrupole time-of-flight mass spectrometry. Multivariate experimental design approaches combined a Minimum-run resolution IV and a Face Centered Central Composite Design were utilized to screen and optimize the DES-USAEME parameters, respectively. Under optimal conditions, the developed method was fully validated, showing low limits of quantification (LOQs; 1.8–10 ng mL^{-1}), satisfactory mean spiked recoveries (74–108%), as well as excellent repeatability and reproducibility precision (2–10%). A preliminary analysis of various samples revealed that 2- hydroxy-benzophenone (2OH-BP) was the most common BP-UVF present in tea beverages.

P87 - Synthesis and application of isotopically labelled reference standards for the mass spectrometric quantification of ergot alkaloids in foodstuff

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Ergot alkaloids form a toxicologically relevant group of mould toxins (mycotoxins) that are among the most common contaminants of foodstuff and animal feed worldwide. Reliable controls are essential to minimise health risks and economic damage. Due to their toxicological relevance, EU limit values for 12 priority ergot alkaloids have been introduced for the first time in 2022 and range from 500 µg/kg in rye milling products down to 20 ug/kg Processed cereal-based foods for infants and young children[1]. High-performance liquid chromatography - mass spectrometry is used to quantify low concentrations of ergots in food, however the European standard analytical procedure cannot be applied due to the lack of isotopically labelled reference standards.

The complex structure of the ergot alkaloids makes a total synthesis extremely challenging, expensive and time-consuming. Therefore, we are focusing on different semi-preparative methods (electrochemistry, organic synthesis, heterogeneous catalysis) to specifically N-demethylate the C8 carbon atom of the lysergic acid moiety. The norergot alkaloid formed is then isotopically labelled using an electrophilic methyl source, i.e. iodomethane or dimethyl sulphate to obtain the specific isotopic labelled ergot alkaloid. Initial experiments have shown that N-demethylation of the ergot alkaloid ergotamine is possible by both electrochemical and wet-chemical organic synthesis. The next step is to improve the previously determined reaction conditions to enable the synthesis of norergotamine on a mg scale for further reactions.

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1. *Commission Regulation (EU) 2021/1399 of 24 August 2021 amending Regulation (EC) No 1881/2006 as regards maximum levels of ergot sclerotia and ergot alkaloids in certain foodstuffs. Official Journal of the European Union*, 2021(L301): p. 1-5.

P88 - A combined MSPE-DLLME-UA methodology for the determination of UV filters and its metabolites in human milk through chromatographic and chemometric resolution

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Organic UV filters are a broad family of compounds of diverse physicochemical characteristics that are present in several cosmetic products aiming to protect users from UV solar radiation. After topic application, UVF can be absorbed through the skin and metabolized in the body and either excreted or bioaccumulated. Notwithstanding the maximum allowable in cosmetic products are strictly regulated, there are no official monitoring protocols for UVF in human fluids and tissues [1]. In this work, we present a sample treatment strategy for the determination of 16 UVF of wide polarity in breast milk with chromatographic and chemometric resolution. The method is based on the combination of a micro solid-phase extraction (MSPE) that allowed isolated polar and non-ionizable UVF, and a dispersive micro liquid-liquid extraction-ultrasound assisted (DLLME-UA) that enabled the extraction of low polarity UVF. The samples were firstly hydrolyzed with β -glucuronidase/aryl sulphatase in an acidic medium and then, an optimized procedure for deproteinization and fat remotion was applied. The samples were eventually subjected to an MSPE-DLME-UA clean-up protocol that enabled the isolation and pre-concentration of the analytes. For the MSPE, the treated samples were passed through Oasis WAX 3 cc Vac cartridge. The experimental conditions of the DLLME-UA were optimized by using a response surface methodology. The samples were subjected to the optimized DLLME-UA which utilizes acetone and acetonitrile as dispersive solvent and trichloromethane as extractant solvent. The final extracts were evaporated and reconstituted in a mixture of ACN:MeOH:H_2O. The samples were analysed through reverse phase chromatographic separation accompanied by UV diode-array and fast-scanning fluorescence detection. The chromatographic analysis enabled the full resolution of 13 analytes, whereas three coeluted analytes were chemometrically resolved by using multivariate curve resolution-alternating least square modelling. With this methodology, a pre-concentration factor of 5 was obtained, reaching limits of detection as low as 1.3 ng mL^{-1} were obtained, and recoveries between 60 % and 100 % for the 16 analytes were accomplished in breast milk samples.

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Reference: Chisvert et al., Analytica Chimica Acta, 752 (2012), 11-29

P89 - Ultra-high sensitivity quantification of veterinary drug residues in products of animal origin

Jianru Stahl-Zeng, Jack Steed, Yoann Fillatre, Daniel McMillan, Michael Scherer, Aline Staub Spörri, Grégoire Bonvin

The use of pharmacologically active substances in veterinary settings has been scrutinized for several years due to their sometimes inappropriate or intensive application. Therefore, these substances must be limited to mitigate negative consequences. One way to implement controls is to perform analytical testing of animal by-products. Several compounds found in these by-products have a maximum residue limit (MRL) to minimize their use, and some compounds are prohibited due to their inherent toxicity. To limit these compounds within the food industry, it is important to achieve LOQ values that are as low as is reasonably possible.

Here, we present ahighly sensitive LC-MS/MS method for analyzing over 180 compounds used in the veterinary industry that can achieve LOQ values as low as 0.005 ng/mL.

As regulations continue to tighten around food testing, it has become increasingly challenging to achieve the necessary levels of sensitivity during analysis while maintaining a high level of accuracy and precision. Within this method, LOQ values down to 0.005 ng/mL have been achieved while keeping high levels of accuracy and precision in standard solutions. When spiked into matrices, LOQs of 0.01 µg/kg in pork and chicken and 0.005 µg/kg in milk were achieved. This high level of sensitivity allows routine laboratories to further dilute their samples to minimize any matrix effects observed. The linear range of each compound analyzed has been assessed, with ranges spanning up to 4 orders of magnitude and r values >0.99. sMRM acquisition helps ensure that both quantifier and qualifier transitions can be measured to increase the specificity of the analysis without the need to compromise on data quality by reducing the number of data points across each peak. In addition, chromatographic separation is important to minimize the number of compounds analyzed at any one time. This allows for a balance between the cycle time of the mass spectrometer and the dwell time for each analyte so that accurate quantification can be performed for each compound.

In addition to MRLs, minimum method performance requirements (MMPRs) are recommended by the EU for some prohibited compounds, which are summarized here. In these instances, the sensitivity of the analysis is paramount to ensure the MMPR is achieved or exceeded. In this method the MMPR has been met or improved upon, showing that even for the most difficult to analyze compounds, this method can achieve or improve on the recommended levels of sensitivity.

An ultra-sensitive and robust method enables the quantification of veterinary drugs using LC-MS/MS in routine laboratories, improving on current regulations.

P90 - A rapid screening and quantitative LC-MS/MS method for food and environmental contaminants using the ZenoTOF 7600 system

Daniel McMillan, Jianru Stahl-Zeng & Robert DiLorenzo SCIEX

Abstract

Pesticides are used before and after harvest to protect crops from infestation by pests and plant diseases. A consequence of their prevalent use in the environment is the appearance of pesticide residues in treated crops and ultimately in human food supplies. Pesticide residues have become a major concern in the area of food safety, and a robust and sensitive screening method for a vast range of pesticides in food matrices is a pressing need. To meet this need, a rapid and robust method was developed for the analysis of food and environmental

contaminants such using the ZenoTOF 7600 system from SCIEX. Zeno trap technology significantly improves sensitivity and electron activated dissociation (EA. D) fragmentation technology improves the accuracy of qualitative analysis results. The ZenoTOF 7600 system is equipped with an EAD cell that simultaneously captures precursor ions and free electrons.

Precursors then form a free radical that dissociates, often generating more fragment ions and a more informative MS/MS, which ensures precise quantification and qualitative analysis for food and environmental contaminant analysis

Novel Aspect:

Zeno trap technology significantly improves pesticide sensitivity, and EAD fragmentation technology improves the accuracy of qualitative results.

P91 - Quantitation of per- and polyfluoroalkyl substances (PFAS) in foodstuffs

This poster addresses the commission regulation (EU) 2022/2388 of 7 December 2022 amending Regulation (EC) 1881/2006 (effective 1st January 2023) regarding the maximum residue levels (MRLs) of 4 per and polyfluoroalkyl substances (PFAS compounds) in certain foodstuffs.[1] Pork liver, honey and egg samples were prepared and spiked by Fera Science Ltd. and instrumental methods were developed to achieve quantitation of per- and polyfluoroalkyl compounds (PFAS) in foodstuffs at the sub µg/kg range. The high sensitivity of the SCIEX 7500 system[2] allowed for a small sample injection volume (2 µL), which is essential to reduce matrix interferences in complex food matricies.

Due to the high sensitivity of the SCIEX 7500 system, only a small volume (2 µL) of food matrix is required, which is beneficial to reduce matrix interference in complex matrices

For PFAS standards in solvent the %CV of area ranged between 1.26 - 14.83% (n=6) with good linearity (r^2>0.99) for each analyte. Therefore, all parameters are within acceptable limits

The calculated concentrations of PFAS compounds that were detected in spiked pork liver samples using the SCIEX 7500 system were found to be in excellent agreement with the results obtained by the EURL proficiency test (PT)

Trace level PFAS contaminants in blank extraction are detected and quantified .

P92 - A fast and novel workflow for screening smoke affected grapes and wine using SPMESH-DART-MS/MS

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Introduction:

Volatile phenols (VPs) are routinely measured via chromatography-based analyses in foods and beverages for quality control. Throughput limitations inherent to chromatography-based approaches can lead to analytical bottlenecks when large amounts of data are required in compressed time frames, i.e. following bushfire events near vineyards. We report translation of a previously described rapid screening method for 5 routinely monitored VPs using solid phase mesh enhanced sorption from headspace-direct analysis in real time-MS/MS (SPMESH-DART-MS/MS) to an automated analysis using an integrated DART-MS system[1].

Objectives:

To demonstrate a simple screening solution for VPs that provides enhanced data quality, faster results, and more cost efficiency as compared to traditional screening approaches.

Methods:

Screening workflow includes the following five VPs (4-ethylphenol (**1**), 4-ethylguiacol (**2**), guaiacol (**3**), 4-methylguaiacol (**4**), and o-cresol (**5**)). Sample preparation (~1.5 hours) was performed for 24 samples in parallel as described in previous work[1]. Following sample preparation and extraction, the SPMESH sheet was transferred to the automated positioning stage of an EVOQ DART-TQ$^+$ mass spectrometer for analysis. MS/MS parameters were optimized (collision energies, collision cell pressure, and scan speed) for translation onto the EVOQ DART-TQ$^+$ system. Matrix matched calibration QC were analyzed, using d_3-guaiacol as IS for all compounds. Regression curves were analyzed at 6 calibration levels in quadruplicate including matrix blanks. Accuracy was assessed using 2 QCs analyzed in quadruplicate. Automated DART-MS/MS analysis of 24 samples was performed in 12 minutes.

Results:

Data processing was performed using standard MS quantitation software for **1-5**. Calibration data for **1-5** was analyzed using linear regression achieving R^2 values of 0.990 or better. Accuracy assessments at 5 µg/L and 25 µg/L were between 90 and 110%. Total workflow time for 24 samples was under 1.5 hours.

Discussion:

This method describes a fully-integrated chromatography-free workflow for screening VPs in foods and beverages by triple quadrupole mass spectrometry. Full workflow simplicity, sample throughput, and data quality meet or exceed the accepted metrics of conventional approaches.

References:

[1]Terry L. Bates, Gavin L. Sacks. Rapid headspace solid-phase microextraction sheets with direct analysis in real time mass spectrometry (SPMESH-DART-MS) of derivatized volatile phenols in grape juices and wines, Analytica Chimica Acta, Volume 1275, 2023.

P93 - Examining the Fatty Acid Profile of the Calf Meat: Investigating the Relationship with Pasture and Food Farming Systems

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This study's primary objective was to examine the relationships between fatty acids and the predominant pattern that distinguishes two distinct agricultural systems based on the presence or absence of graze practices. In particular, the study assessed the effect of grazing on the fatty acid composition of meat samples collected from two farms during the 2022 season. It is essential to note that these farms were meticulously chosen to ensure that their non-grazing season locations and management practices were identical. The concentrations of fatty acids were determined using gas chromatography, a reliable analytical technique [1]. The obtained results demonstrated significant differences in the fatty acid compositions of meat samples from grazing and non-grazing agricultural systems [2]. These results suggest that the presence or absence of grazing practices has a discernible effect [3] on the fatty acid composition of calf meat. By expanding the scope of this study and examining the differences in fatty acid content between the two agricultural systems, we can obtain valuable insights into the potential impact of grazing practices on the nutritional characteristics of meat. These results may have significant ramifications for producers, consumers, and the agricultural industry as a whole. Further analysis and investigation of this subject could contribute to the development of environmentally responsible and sustainable agricultural practices that maximize the nutritional value of livestock products.

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P94 - Determination of Arsenic Species in Rice from Different Countries Using High-performance Liquid Chromatography – Inductively Coupled Plasma – Mass Spectrometry

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\In recent years, increased attention has been paid to arsenic due to its carcinogenic properties and other toxic effects. There is worldwide concern regarding the inorganic arsenic contamination of rice due to its associated risks. According to the International Agency for Cancer Research (IRAC), arsenic (As) is a human carcinogen, and even small amounts of it can prove harmful. There are two types of arsenic compound: organic and inorganic (iAs), with the later form, considered the most toxic. Because of different toxicity and chemical properties are shown depending on their oxidation states and elements, studying the speciation of arsenic is as important as quantifying its total quantity. In the present study, the main objectives were to examine the total arsenic and arsenic speciation levels in different types of rice using inductively coupled plasma/mass spectroscopy (ICP–MS) and high-performance liquid chromatography (HPLC–ICP/MS). This experiment focused mainly on detecting four arsenic species; namely, arsenite (AsIII), arsenate (AsV), monomethyl arsenic acid (MMA), and dimethyl arsenic acid (DMA). Twenty-eight types of rice samples were collected from Saudi markets, that had been imported from different countries. All of the samples of polished rice were analysed, and the results were compared to the maximum FAO/Codex limit. To prepare the samples, the rice was mixed with a 0.28 M HNO_3 solution and heated at 95 °C for 90 minutes, then the extracts were initially diluted with deionized water, centrifuged, filtered, and then diluted further, while adjusting the pH level prior to analysis using HPLC-ICP-MS. The spiked sample percentage recovery for iAs, DMA, and MMA ranged from 85-110.4%. The results showed that the mean for the total arsenic, inorganic and organic arsenic levels in the rice samples were 109.40±48.39, 76.28±29.72 and 33.12±18.29 ng/g, respectively (n= 28). The level in the inorganic As were higher than organic As in all samples; the major detection of inorganic arsenic was arsenate (AsIII), while in the organic arsenic, only trace levels were found. It was noted that the samples imported from Italy possessed the highest concentration, and one exceeded the maximum level.

From our findings, we can conclude that, based on the high AsIII predominance in rice grains, there is a toxic effect of As in rice, which needs to be explored further in order to determine the cause of these toxic effects and review the current maximum limit for mitigating its impacts on human health

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P95 - Determination Of Pesticide Residues In Cucumber Using Gc-Ms/Ms With Apgc

JD de Alwis, Stuart Adams, Doug Stevens
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P96 - Determination of tropane alkaloids in cereal-based baby food and vegetables: the case study of the Italian contaminated spinach

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Tropane alkaloids (TA) are toxic compounds produced by several plants, such as *Datura stramonium* and *Atropa belladonna* and other weed plants that often grow among food crops. The co-harvesting of these plants with the crops can cause TAs contamination in food and for that reason the EU set maximum limits in baby food, some cereals and derived products, herbal infusions [1] and recommended monitoring activities. Nevertheless, two cases of consumers intoxication due to the presence of TA-producing weed plants in commercially available spinach highlighted the concern about TA presence in food not included in the official controls. To face the health emergency, a test method to determine atropine and scopolamine was developed and validated, although the absence of an EU maximum limit set for vegetables. The EURL method for atropine and scopolamine determination in cereal-based baby food [2] was modified to be applied also to leafy vegetables. Method performance parameters were evaluated for both TAs by spiking blank samples of spinach and broccoli, biscuits and creams for infants and babies at 0.3 (LOQ) and 1.0 µg/kg. The LC-MS/MS analysis was carried out on an ExionLC system coupled to a QTRAP 6500$^+$ mass spectrometer, in MRM mode. After validation, the method was applied to determine atropine and scopolamine in some cereal-based baby food and leafy products. We tested 17 samples of cereal-based baby food collected from the market; 3 out of 17 (the 17.6%) were contaminated by atropine between 0.4 and 1.8 µg/kg. The spinach that caused consumer's intoxication did not contain the two TAs, but the weed plant seed found in it had scopolamine between the LOD and the LOQ. The seed capsule was identified as belonging to the *Xanthium L.*, a widespread infesting plant in Italian crops. Since the chromatograms of both the seed capsule and the spinach showed some matching peaks of the scopolamine MRM transitions but at a different retention time, we suspected the presence of other TAs that could have caused the intoxication. To annotate the unknown TA and other toxic plant metabolites, we analyzed the seed extract using a tribrid Orbitrap IQ-X high resolution mass spectrometer, performing untargeted MS acquisition. The features were annotated against on-line databases and level I confirmation [3] with analytical standard is undergoing.

1. Commission Regulation (EU) 2023/915 of 25 April 2023.
2. EURL-MP-Method_004 (version 1).
3. DOI: 10.1021/es5002105

P97 - Glycoalkaloids in (processed) table and industrial potatoes

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Text: Glycoalkaloids (GAs) can be present in high amounts in potatoes, causing adverse effects in animals and humans. The main GAs in potato are α-solanine and α-chaconine, consisting of the same aglycone, solanidine. Upon food processing, α-solanine and α-chaconine may be partially or completely degraded to β- and γ- forms or solanidine, respectively. According to BfR, the total GA content in potatoes should not exceed 100 mg/kg fresh weight (f.w.) [1]. Sweden has also established a legal limit of 200 mg/kg f.w. for table potatoes [2, 3]. In 2022, European Commission published Recommendation 2022/561 to monitor α-solanine and α-chaconine in potatoes and their products [4]. It was recommended that all degradation products of solanine and chaconine should be analyzed and the limit of quantification (LOQ) for individual GAs to be around 1 mg/kg [4]. To determine the occurrence of GAs in (processed) table and industrial potatoes, a straightforward method was developed and validated [5]. The GAs (α-solanine, α-chaconine, γ-chaconine and solanidine) were extracted from fresh and processed (peeled and cooked) "waxy" and "floury" varieties of potatoes with a mixture of methanol/water/formic acid and analyzed by LC-MS/MS. Quantification was performed by external calibration in blank surrogate matrix extract. The LOQs obtained were 0.5 mg/kg for α-solanine and α-chaconine and 0.1 mg/kg for γ-chaconine and solanidine. The highest concentration of total GAs was found in raw whole potatoes of a floury variety at 221 mg/kg f.w. and two samples (of waxy and floury varieties) with GAs over 100 mg/kg f.w., all exceeding the limits set by Sweden and BfR, respectively. The examined raw whole potato samples contained on average 70.7 mg/kg of GAs. Peeling removed on average 78.1% of the total GAs from raw potatoes, 84.3% from waxy and 67.7% from floury varieties. None of the peeled samples contained GAs above 100 mg/kg. Boiling of potatoes further decreased GAs by 7.1%. Overall, peeling and boiling reduced total GAs by 85.1% on average. However, high percentages of GAs in raw peeled potato compared to raw unpeeled potato were found in three floury cultivars ranging from 47.3 - 88.4% and in one waxy cultivar, being 41.3%. This suggests that some potato cultivars contain relatively high levels of GAs in the potato flesh.

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4. EU, *THE EUROPEAN COMMISSION, COMMISSION RECOMMENDATION (EU) 2022/561 of 6 April 2022 on monitoring the presence of glycoalkaloids in potatoes and potato-derived products.* Official Journal of the European Union, 2022. **L 108**: p. 66-67.
5. EURL, *EURLMP-method_014 v1, 2023, Determination of glycoalkaloids in potatoes by LC-MS/MS.* 2023.

P98 - Matrix effects in the analysis of high polarity pesticides in feed and cereal products using LC-MS/MS

List of authors: A. Perczak[1], A. Nowacka[1], A. Hołodyńska-Kulas[1], F. Stachowiak[1], D. Petlińska[1], D. Drożdżyński[1]
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Text: The analysis of high polar pesticides is a challenge for departments involved in the monitoring of pesticides in plant origin products. It is an uncomplicated extraction procedure, which results in the occurrence of many undesirable water-soluble chemical compounds from various plant matrices in the final samples. It affects the reduction of signals from LC-MS/MS, which causes irregularities in quantitative analyses. Several different grain-feed matrices were used in the research, which were added to analytical standards and then the matrix effects were checked against standards prepared in solvents.

Five grain-feed matrices were used in the study: wheat, buckwheat, soybean, oat and barley. Extracts from products that did not contain the analyzed active substances were prepared using the QuPPe extraction method [1] - water/acidified methyl alcohol mixture was added to the samples, then they were shaken and frozen. After centrifugation, the sample was filtered into polypropylene tubes and then added to chromatographic bottles containing the appropriate concentration of the mixture of standards (finally 1, 5, 10, 20 and 50 ng/ml), which included: amitrole, AMPA, chlormequat, cyromazine, ethephon, fosetyl-Al, glyphosate, glufosinate, melamine, mepiquat, MPPA, N-acetyl-AMPA, N-acetyl-glyphosate, N-acetyl-glufosinate. The analysis was performed using a highly sensitive LC-MS/MS apparatus (SCIEX Triple Quad 7500 LC-MS/MS System).

Depending on the type of active substance and the used matrix, large diversity in matrix effects was observed. The greatest matrix effects, in relation to standards prepared in pure solvents, were noted for amitrole, melamine, cyromazine and AMPA, while for such active substances as chlormequat, ethephon, glyphosate, mepiquat this effect was not observed or it was minimal. The matrix that resulted in the lowest intensities compared to the other experiments was soybean. In turn, the greatest differentiation in matrix effects was observed in the analysis of fosetyl-Al.

High variation in matrix effects was noted. If this effect is not observed, it will be possible to perform analyzes without using internal standards and calibration curves prepared in matrix extracts, while with high effects, the above actions should be taken in order to perform an appropriate quantitative analysis of plant protection products.

References:

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P99 - Fully automated workflow for Sample Preparation and Measurement of Dioxin in Food and Environmental samples

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Dioxins are a class of very toxic compounds found throughout the world in the environment. Equipment sensitivity is of great importance for the analysis of low concentrations of these highly-toxic compounds. Historically, analysis and detection of dioxins was done with magnetic sector-type high-resolution mass spectrometers (HRMS). However, in recent years, the performance of triple quadrupole mass spectrometers (MS/MS) has improved significantly. In addition, the development of the Boosted Efficiency Ion Source (BEIS) offers compound-specific sensitivity up to 4 times greater than previous ion sources and provides accurate quantitation of dioxins at levels comparable to HRMS. Detection limits as low as 20 fg for Tetrachlorodibenzo-p-dioxin (TCDD) were achieved. In this study, we analyzed dioxins in about 250 samples of approximately 40 types of food and animal feed products using a GC-MS/MS with BEIS. We also evaluated the number of analyses possible while maintaining sensitivity at low concentrations in order to verify the durability of the GC-MS/MS instrument.

P100 - Determination of Alternaria toxins in food by HPLC-MS/MS

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[1]Eurofins WEJ Contaminants GmbH, Hamburg, Germany

Alternaria toxins belong to the group of mycotoxins and are formed naturally by certain fungi of the genus *Alternaria* (black mould). Due to their ubiquitous distribution, these toxins can be found in many food groups such as cereals, tree nuts, vegetables (tomatoes, carrots, potatoes), fruits (apples, grapes), oilseeds (sunflower seeds, canola), olives, and spices (paprika powder, chilies). Humid climates in the summer months can cause the fungi to proliferate, increasing the risk of contamination with Alternaria toxins. COMMISSION RECOMMENDATION (EU) 2022/553 [1] lists indicative levels for alternariol (AOH), alternariol monomethyl ether (AME) and tenuazonic acid (TEA) for certain foods, based on monitoring results from the European Food Safety Authority (EFSA), above which investigations should be performed. Further relevant Alternaria toxins are altenuene (ALT) and tentoxin (TEN).

A method has been developed for the fast and cost-efficient determination of the 5 relevant Alternaria toxins and validated for various food matrices. Since the distribution of mycotoxins is not homogeneous, a slurry is prepared from certain foods such as spices, nuts, dried fruits, and products made from them. The samples are prepared by an acidic salt-assisted liquid-liquid extraction (SALLE) without any further clean-up or enrichment step. The measurement is performed via high performance liquid chromatographic-tandem mass spectrometry (HPLC-MS/MS). The analytes are separated using an Agilent InfinityLab Poroshell 120 EC-C18 column and LC-MS eluents with a pH of 7.8. Limits of quantitation (LoQ) below the indicative levels are achieved for all specified matrices. For example, LoQ of 0.8 µg/kg for AOH and AME and 2 µg/kg for TEA are reached in infant cereal-based foods with an accuracy of 70-120% and a coefficient of variation of <13%.

[1] COMMISSION RECOMMENDATION (EU) 2022/553 of 5 April 2022 on monitoring the presence of Alternaria toxins in food

P101 - Risk assessment of pharmaceuticals and microplastics in irrigated crops with reclaimed water in Almeria, south East of Spain

Amadeo R. Fernandez-Alba and M. Jesus Bueno
University of Almeria, Spain

The reuse of reclaimed water contributes to mitigating water stress and represents a sustainable alternative for entrepreneurs and farmers in areas where there are large commercial interests and which suffer from greater water scarcity, as is the case of Almeria. However, hundreds of scientific studies have documented the presence of organic contaminants of emerging concern (CECs), such as pharmaceuticals, in reclaimed water and irrigation water. In addition, new contaminants, plastic waste, are posing a new challenge to the scientific community, due to their increasing presence in the environment and their potential negative impacts on ecosystems and human health. An assessment of the possible risks derived from the use of reclaimed wastewater for agricultural irrigation of protected greenhouse horticultural crops in the province of Almeria is of vital importance.

Three crops (cucumber, pepper and melon) were grown under agronomic conditions in a greenhouse. The plants were irrigated with treated water spiked with 70 chemicals, including antibiotics, anti-inflammatories, analgesics, anaesthetics, anxiolytics, and anticonvulsants at environmental concentrations (~ 1 µg/L). The average total concentration of contaminants detected in the soil samples ranged from 106 to 217 µg/kg d.w depending on the crops type. Melon roots showed a higher number of contaminants than pepper and with mean total concentrations ranged from 196 to 635 µg/kg. Between 5 and 12 different contaminants were found in the harvested fruits, up to levels around 23 µg/kg in cucumber and pepper, and 13 µg/kg in melon. In general, cucumber fruit exhibited accumulation percentages of contaminants higher than the other fruits (pepper and melon) for most target analytes. The accumulation rates followed the order: root = stem (0.1-0.4%) < leaf (1-3%) < fruit (0.2-5%) < soil (13-28%). The experimental data obtained in this study were also used to assess the risk associated with the reuse of reclaimed water for crop irrigation as well to identify those contaminants that, due to their physicochemical properties and accumulation percentages may pose a greater environmental impact. Carbamazepine and lidocaine showed the highest translocation rate through the xylem plant tissue and accumulation percentages in the leaves. Clonazepam and carbamazepine were the contaminants that showed the highest bioconcentration factor values (BCF) in vegetables. The total load of contaminants detected derived from the consumption of a mix of all vegetables were up to 8 orders of magnitude lower than the acceptable daily intake values (ADI) reported in the worse of the case, 380 ng/day for a conventional or 1145 ng/day for a vegetarian diet.

P102 - Retrospective hrMS analysis of veterinary drugs and growth promoters in bovine urine

E. de Lange, M.G.M. van de Schans, E. J. Jongedijk, A. Lommen, M.H. Blokland

Wageningen Food Safety Research, part of Wageningen University and Research

The current mass spectrometric methods used for food safety monitoring are generally based on targeted measurements. Therefore, any compounds outside this predefined list of compounds cannot be measured. In the current situation, compounds need to be added to the method's target list in case of a new chemical hazard or the addition of previous unknown marker metabolites. High-resolution mass spectrometry (HRMS) measurements are not limited to such a predefined targeted list of compounds. Therefore, HRMS is becoming an important technique for food safety monitoring, including multi-class screening methods for a broad range of compound groups [1, 2], identification of new emerging compounds [2], metabolite studies [2-4], and retrospective analysis [2, 5]. In this study, we focused on the retrospective analysis of HRMS data. In our new approach, we generated an LC-HRMS data archive of bovine urines from 2015 to 2022. The data files were processed with our in-house developed software suite MetAlign. The data processing was performed in two steps: 1) reducing the 2968 LC-HRMS data files 100-fold in size, including a correction of the exact mass and retention time, and 2) ultra-fast searching of the reduced data files for 181 growth promoters and 136 veterinary drugs. We will present this retrospective workflow and two proofs of concept:

1) Occurrence of growth promoters and veterinary drugs in bovine urine
2) Trend analysis of antibiotic occurrence per year, in which the results are compared with antibiotic sales data

Literature

1. Wang, H., et al., *Screening and quantification of 146 veterinary drug residues in beef and chicken using QuEChERS combined with high performance liquid chromatography-quadrupole orbitrap mass spectrometry.* Food Chemistry, 2023. **408**: p. 135207.
2. Jongedijk, E., et al., *Use of high-resolution mass spectrometry for veterinary drug multi-residue analysis.* Food Control, 2023. **145**: p. 109488.
3. Gómez-Pérez, M.L., et al., *Identification of transformation products of pesticides and veterinary drugs in food and related matrices: Use of retrospective analysis.* Journal of Chromatography A, 2015. **1389**: p. 133-138.
4. Huber, C., et al., *A large scale multi-laboratory suspect screening of pesticide metabolites in human biomonitoring: From tentative annotations to verified occurrences.* Environment International, 2022. **168**: p. 107452.
5. Kaufmann, A., et al., *Potential and limitation of retrospective HRMS based data analysis: "Have meat-producing animals been exposed to illegal growth promotors such as SARMs?".* Food Control, 2023. **147**: p. 109611.

P103 - Analyzing of Zilpaterol Hydrochloride in red meat samples available in Saudi markets by HPLC-MS/MS

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[1] Reference Laboratory for Food Chemistry, Saudi Food & Drug Authority (SFDA),
Riyadh 11561, Saudi Arabia.

Zilpaterol Hydrochloride is a growth promoter product that belongs to the class of beta-adrenergic receptor agonists. The use of Zilpaterol Hydrochloride has been demonstrated to improve lean carcass characteristics, increase growth potential, and decrease feed intake in cattle and lambs. Although Zilpaterol Hydrochloride has been approved for use in feedlot cattle in Brazil, Canada, South Korea, and Mexico, it has been banned in several countries, including China and the European Union. Due to concerns that zilpaterol may pose a health risk to humans, Zilpaterol Hydrochloride is not currently used in any feeding system in the United States. As a result of insufficient information, some countries have restricted or banned the import of meat that contains traces of Zilpaterol Hydrochloride due to concerns regarding human health. This study aimed to analyze the Zilpaterol Hydrochloride in 13 red meat samples were purchased from Saudi markets. The extraction method of these samples was based on the QuEChERS followed by LC- MS/MS Analysis. In the LC-MS/MS method, the recovery value ranges from 99 to 108.4

%, with a RSD of 5.13 %. There is satisfactory linearity in the calibration curves, with a coefficient of determination (R^2) > 0.999 for the analyte. Fortunately, all analyzed samples were below LOQ, which means the investigated products are safe for human consumption. Moreover, our results reflect the efficiency of the SFDA's regulations for monitoring and controlling foods in Saudi markets.

References:

1. Cayetano-De-Jesus, Jorge Adalberto, Rolando Rojo-Rubio, Alicia Grajales-Lagunes, Leonel Avendaño-Reyes, Ulises Macias-Cruz, Veronica Gonzalez-del-Prado, Agustin Olmedo-Juárez, Alfonso Chay-Canul, José Alejandro Roque-Jiménez, and Héctor Aarón Lee-Rangel. "Effect of zilpaterol hydrochloride on performance and meat quality in finishing lambs." Agriculture 10, no. 6 (2020): 241.

Caetano, Mariana, Rodrigo Silva Goulart, Saulo Luz Silva, Sergio Bertelli Pflanzer, Paulo Roberto Leme, Antonio Carlos Ramos Dos Santos, and Dante Pazzanese Duarte Lanna. "Meat Quality of Nellore Young Bulls—Effects of Different Days on Feed and Zilpaterol Hydrochloride Supplementation." Animals 11, no. 9 (2021): 2688.

P104 - An ultra-high sensitivity and robust analysis of PFAS compounds in multiple water sources.

Bertram Nieland, Jack Steed, Rens-Jan Jaspers, Kay Hup, Abdessamad Chahboun, Jianru Stahl-Zeng, Daniel McMillian

PFAS compounds are becoming increasingly ubiquitous in our environment due to overuse and their lack of breakdown, ensuring that this will be a challenge for decades to come. Therefore, it is imperative to provide rigorous and sensitive analytical testing to regulate these compounds and try to limit their possible effects on human health

As the limits for PFAS compounds continue to be reassessed and reduced it becomes increasingly important to provide methods which achieve limits of detection which are as low as is reasonably possible

It has been well documented that when performing PFAS analysis contamination and interference can be large challenges of a successful analysis due to the numerous sources of these compounds and the interferences which can be observed. Here, we detail the steps which we recommend being taken to ensure the analysis of PFAS compounds is as simple and challenge free as possible:

Overall, it is possible to achieve ultra-high levels of sensitivity for PFAS compounds down to 0.2 ng/L when using the SCIEX 7500 System. Excellent levels of precision, accuracy and linearity have been achieved for all compounds analyzed. Detection limits down to 0.06 ng/L in three different water matrices. The importance of reducing interferences and contamination is paramount to a successful PFAS analysis. And long term stability of the method will be demonstrated in this poster.

P105 - A green approach to Total Petroleum Hydrocarbon (TPH) Analysis according to "ISO 9377-2:2000. Water quality — Determination of hydrocarbon oil index" and "U.S. EPA 5021A (2003), "Volatile Organic compounds in soil and other matrices using equilibrium headspace analysis": complete automation and massive solvent saving

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Abstract

The TPH quantitation in water sample is regulated in many EU State members. It involves the analysis of two hydrocarbon fraction: VPH (also called C<12) and EPH (also called C10-C40). In particular, Italian regulation asks for several water matrices an LOQ in the range of tenth ppb. The reference method for EPH (ISO 9377-2) requires one liter of sample and more than 50 ml of hexane, not to mention a high amount of Florisil ® and sodium sulphate. Furthermore, the sample preparation requires lot of operator time, as well as carefully cleaned glassware. The solution proposed by SRA is able to save up to 90% of the needed volume of hexane and reduces up to 1/50 the sample volume, as well as the amount of Florisil®. The whole sample prep is totally automated, with no evaporation step which can cause loss of volatile fraction. The operator is only asked to place samples on autosampler and start the process.

In addition, the analytical platform is suitable to run both determinations (VPH and EPH) without any operator attendance.

References:

1- ISO 9377-2:2000. Water quality — Determination of hydrocarbon oil index — Part 2: Method using solvent extraction and gas chromatography.
2- Grob, K. (1987). On-column injection in capillary gas chromatography: Basic technique, retention gaps, solvent effects. Hüthig Verlag Heidelberg. ISBN 3-7785-1551-9.

P106 - Predicting acrylamide content in vegetable crisps using a rapid and non-destructive smartphone-based method

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Acrylamide (2-propenamide CAS 79-06-01, MW 71.08 g/mol) is a thermal processing contaminant commonly found in heat treated foodstuffs (> 120 °C) of plant origin containing reducing sugars and asparagine as crucial precursors. Importantly, although acrylamide has been classified as a probable human carcinogen (Group 2A), it is not legislatively regulated and only benchmark levels have been set in the EU (Regulation 2017/2158). In case of benchmark level excess, mitigation measures should be taken, e.g., milder thermal processing. However, there is an ongoing discussion on establishing maximum limits for acrylamide indicating the need to develop methods for its accurate and sensitive detection [1]. Acrylamide is commonly measured by instrumental methods, such as liquid chromatography coupled to mass spectrometry (LC-MS), nevertheless, alternatives featuring simplicity, cost- effectiveness and rapidness have been also proposed. In detail, acrylamide formation has been correlated with the formation of dark colour on the processed food surface, and actually the darker the emerging colour the higher the acrylamide concentration [2]. In this study, to achieve non-destructive and cost-effective acrylamide screening, we utilized a smartphone as an optical detector to monitor the colour of vegetable crisps prepared from carrot, purple carrot, yellow carrot and sweet potato. Two different thermal treatments were applied, namely, conventional and vacuum frying, and kinetic measurements with 3 endpoints were performed in both cases. Considering the different colour matrix on each occasion, an image data processing protocol was applied and the RGB colour space was used as the analytical signal. It was found that the R values correlated the best towards the LC-MS data in all cases. The only exception was in the case of purple carrot, in which no correlations could be obtained, as its naturally dark colour hindered the formation of visually significant changes. All in all, this study highlights the integration of smartphones into chemical analysis, significantly reducing the analysis cost and potentially enabling at home analysis.

References

[1] European commission, Standing Committee on Plants, Animals, Food and Feed Section Novel Food and Toxicological Safety of the Food Chain, Summary report on 27/2/2023

[2] R. Sáez-Hernández et al., Determination of acrylamide in toasts using digital image colorimetry by smartphone, 2022, Food Control, 109163

P107 - River epilithic biofilms, POCIS, and water samples as complementary sources of information for a more comprehensive view of aquatic contamination by pesticides and pharmaceuticals

R. Zanella[1], D.R. Dos Santos[1], M.C. Bastos[1,2,3], J.A.M.De C. Lima[1], T. Le Guet[2], J.V. Brunet[1,3], G. Fernandes[1], O.D. Prestes[1], L. Mondamert[3] and J. Labanowski[3]
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Spatial-temporal monitoring of the presence of pesticides and pharmaceuticals in water requires an adequate evaluation of different sampling strategies for the choice of the matrix to be analyzed. The use of matrices, isolated or combined, may better represent the real state of contamination by organic compounds. In this sense, the present work compared the effectiveness of using river epilithic biofilms with active water sampling and with the passive sampler POCIS. Analysis were performed by ultra-high performance liquid chromatography with tandem mass spectrometry (UHPLC-MS/MS). A watershed representative of South American agriculture was monitored. Nine sites with different rural anthropic pressures (natural forest, intensive use of pesticides, and animal waste), and urban areas without sewage treatment, were monitored. Water and epilithic biofilms were collected during periods of intensive pesticide and animal waste application. After the harvest of the spring/summer crop, a period of low agrochemical input, the presence of pesticides and pharmaceuticals was monitored using the POCIS and epilithic biofilms. The spot water sampling leads to underestimation of the level of contamination of water resources as it does not allow discrimination of different anthropic pressures in rural areas. The use of endogenous epilithic biofilms as a matrix for the analysis of pesticides and pharmaceuticals is a viable and highly recommended alternative to diagnose the health of water sources, especially if associated with the use of POCIS.

References:

D.R. Dos Santos, M.C. Bastos, J.A.M.De C. Lima, T. Le Guet, J.V. Brunet, G. Fernandes, R. Zanella, O.D. Prestes, L. Mondamert, J, Labanowski (2023) Journal of Environmental Science and Health, Part B, 58:3, 273-284, DOI: 10.1080/03601234.2023.2182583.

Vendor Seminar Presentations

Advances in MS technology for food analysis: SCIEX 7500 for PFAS; introduction to ZenoTOF 7600

Jianru Stahl-Zeng and Daniel McMillan

Food, Environmental and Forensics (EMEAI), SCIEX

Workshop presentations will include:

- The benefits of industry-leading sensitivity and robustness with the SCIEX 7500 system for PFAS analysis
- An introduction to the ZenoTOF 7600 system for advanced workflows in food research

An overview of recent application examples

Recent developments of the µSPE clean-up workflow in pesticides analysis

Hans-Joachim Hübschmann, Thomi Preiswerk
CTC Analytics AG, Zwingen, Switzerland

Microextraction techniques in sample preparation have emerged as alternatives to traditional extraction techniques. Classical standard methods can be successfully scaled down to miniaturized methods. With the scale-down to only microliter volumes successful automation of these established methods is the next logical step. In contrast to only automate a former manual procedure, for instance by using the classical large volume SPE cartridges and replacing the manual steps, for a true green analytical chemistry, a new µSPE cartridge was introduced for automated processing of raw extracts for online or offline pesticides analysis by GC-MS or LC-MS. The use of this mini cartridge in the fully automated µSPE clean-up workflow did show its versatility for pesticides analysis cleaning-up extracts from acetonitrile (ACN) or ethylacetate (EtOAc).

Daily Conference Program

Monday 20 November

Date	Time	Title	Room
Mon 20 Nov	13:00-17:00	AOAC Workshop: Best Practices for Bioassay Testing of Food and other Complex Mixtures	Calla 1
	12:00-18:00	Registation	Lobby
	12:00-18:00	Poster setup	Exhibition room
	17:30-19:00	Welcoming drinks	Exhibition room

Tuesday 21 November

Time	Session	Title	Presenter	Organisation	Room
8:00-9:15	**Registration & poster setup**				Registration desk
8:45-9:00	**Opening Ceremony**				Plenary
	Keynotes (Chairs: Marja Lamoree & Hans Mol)				Plenary
9:00-9:30	The present and future use of monitoring data in EFSA's human health risk assessment of chemicals		Bruno Dujardin	EFSA	Plenary
9:30-10:00	From target to non-target screening of chemical mixtures in water - in situ, frequently, hazard-based, retrospectively		Juliane Hollender	Eawag	Plenary
10:00-10:15	Keynote Q&A				Plenary
10:15-11:00	**Exhibition, Posters & Coffee**				Exhibition room
	Plenary session - Suspect and non-target screening (Chairs: Nestor Etxebarría & Laura Righetti)				Plenary
11:00-11:20	Long-term targeted and non-targeted monitoring of drinkable water treatment plants in Biscay (Basque Country)		Nestor Etxebarria	University of the Basque country (UPV/EHU)	Plenary
11:20-11:40	Structural alerts for prioritization of potentially toxic substances in non-target screening		Nienke Meekel	KWR/Vrije Universiteit Amsterdam	Plenary
11:40-12:00	Two-stage LC/HRMS workflow for identification of effect drivers in passive sampler extracts after their selective recovery by pull-down assay.		Roman Grabic	University of South Bohemia	Plenary
12:00-12:20	Determination of real-life environmental and food chemical mixtures in water, fish and milk samples using suspect screening coupled to iterative data-dependent analysis		Solene Motteau	Oniris	Plenary
12:20-12:40	Identification of emerging organic contaminants in greywater emitted from ships by a comprehensive LC-HRMS target and suspect screening approach		Meritxell Gros	Catalan Institute for Water Research	Plenary
12:40-13:30	**Lunch in restaurant**				Restaurant
	Plenary session - Suspect and non-target screening (Chairs: Lubertus Bijlsma & Frederic Béen)				Plenary
13:30-13:50	Trend analysis of non-target screening features in surface water for safe drinking water production		Geert Franken	Het Waterlaboratorium	Plenary

Tuesday 21 November

Time	Session	Title	Presenter	Organisation	Room
13:50-14:10	Decoding the human exposome: monitorization of chemicals of emerging concern and their phase II metabolites in urine by target, suspect and non-target screening		Mikel Musatadi	University of the Basque country (UPV/EHU)	Plenary
14:10-14:30	Using LCMS Analysis and Suspect and Non-target Screening to Assess the Efficiency of Hybrid Electrodialysis-Forward Osmosis (ED-FO) in Water Reuse		Manuel Garcia-Jaramillo	Oregon State University	Plenary
14:30-14:50	Data science enhanced non-targeted analysis for characterization of exposome: move beyond structural elucidation		Saer Samanipour	University of Amsterdam	Plenary
14:50-15:30	**Exhibition, Posters & Coffee**				Exhibition room
	Parallel session - Food fraud & authenticity (Chairs: Hans Mol & Laura Righetti)				Calla 1
15:30-15:50	Ultra-rapid, reliable, non-targeted authentication of monofloral honey by dielectric barrier discharge ionization high resolution mass spectrometry (DBDI-HRMS)		Alessandra Tata	Istituto zooprofilattico sperimentale delle Venezie	Calla 1
15:50-16:10	Food Authentication: Mass Spectrometric Strategies for Detecting Food Fraud		Marina Creydt	University of Hamburg	Calla 1
16:10-16:30	Stable Isotope Ratio Analysis for the Authentication of Organic Wheat and Evaluation of Mycotoxin Contents and Residue Levels: From Milling to Pasta and Bakery Products		Zoe Giannioti	Fondazione Edmund Mach	Calla 1
16:30-16:50	Spatial and temporal biodiversity of grape varieties from the Portuguese Bairrada Appellation: in the path of varietal patterns		Sílvia Rocha	Universidade de Aveiro	Calla 1
	Parallel session - Toxic chemicals, metabolites and transformation products (1) (Chairs: Tito Quintana & Marja Lamoree)				Calla 2
15:30-15:45	Estimation of antibiotics usage and resistance genes prevalence using Wastewater-Based Epidemiology as surveillance tool		Lubertus Bijlsma	University Jaume I	Calla 2
15:45-16:00	Wastewater-based epidemiology: a comprehensive public health surveillance tool		Nicola Ceolotto	University of Bath	Calla 2
16:00-16:15	Evaluation of antibiotic levels in a real water reuse system for agricultural irrigation		Patricia Plaza-Bolaños	University of Almeria	Calla 2
16:15-16:30	Investigation of pesticides and pharmaceuticals in small water bodies using polar organic chemical integrative samplers (POCIS) and non-target screening		Ivo Havranek	Norwegian Institute for Bioeconomy Research	Calla 2
16:30-16:45	Tracking organic pollutants and identifying sources in urban groundwaters		Sandra Pérez	IDAEA-CSIC	Calla 2
16:45-17:00	Sample preparation investigation to extract plastic additives in agricultural soil using UHPLC-MS/MS		Aristeidis Tsagkaris	UCT Prague	Calla 2
17:00-17:30		**Break**			Exhibition room
17:30-18:15		Keynote - Partnership for the Assessment of Risks from Chemicals (PARC) (Chair: Marja Lamoree)	Jean-Philippe Antignac	INRAE	Plenary

Wednesday 22 November

Time	Session	Title	Presenter	Organisation	Room
	Keynotes (Chairs: Frederic Béen & Laura Righetti)				Plenary
9:00-9:30		Chemical cocktails threaten the environment and human health: it's all in the mix!"	Beate Escher	UFZ	Plenary
9:30-10:00		Advances in microplastic exposure, fate, effects and risks	Bart Koelmans	Wageningen University and Research	Plenary
10:00-10:15		Keynote Q&A			Plenary
10:15-11:00	**Exhibition, Posters & Coffee**				Exhibition room
	Plenary session - Risk assessment and modelling (Chairs: Montserra Filella & Frederic Béen)				Plenary
11:00-11:15		PERK: A Cutting-Edge R-Based Tool for Enhanced Environmental Risk Assessment of Active Pharmaceutical Ingredients in Wastewater	Kishore Kumar Jagadeesan	University of Bath	Plenary
11:15-11:30		Elimination resistant: Modelling multi-compartment toxicokinetics of thiacloprid in Gammarus pulex using monitoring data, bioconcentration and receptor binding assays	Johannes Raths	Eawag	Plenary
11:30-11:45		Prediction of Acute Fish Toxicity using Molecular Fingerprints and Machine Learning	Viktoriia Turkina	University of Amsterdam	Plenary
11:45-12:00		Agglomeration behaviour and fate of food-grade titanium dioxide in human gastrointestinal digestion and in the lysosomal environment	Francesca Ferraris	Italian National Institute of heath	Plenary
12:00-12:15		In-vitro digestion approach for assessing the bioaccessibility of pharmaceuticals in seafood	Lúcia Santos	Catalan Institute for Water Research (ICRA)	Plenary
12:15-12:30		Urine concentrations of Arsenic and its species in adolescents from Wallonia (Belgium).	Heidi Demaegdt	Sciensano	Plenary
12:30-13:30	**Lunch in restaurant**				Restaurant
13:30-14:00	Vendor session	Advances in MS technology for food analysis: SCIEX 7500 for PFAS; introduction to ZenoTOF 7600	Jianru Stahl-Zeng, Daniel McMillan	Sciex	Calla 2
14:00-14:30	**Exhibition & Posters**				Exhibition room
14:30-15:00	Vendor session	Recent Developments of the µSPE Clean-up Workflow in Pesticides Analysis	Thomas Preiswerk	CTC Analytics	Calla 2
15:00-15:30	**Exhibition, Posters & Coffee**				Exhibition room
	Parallel session - Environmental contaminants & residues in food (Chairs: Amadeo Fernández-Alba & Hans Mol)				Calla 1
15:30-15:45		Fate of PFAS from soil to crop: understanding uptake and how emissions from a PFAS factory in The Netherlands impact levels in fruits and vegetables	Stefan Van Leeuwen	Wageningen University and Research	Calla 1
15:45-16:00		Combined action of ion mobility (IM-MS) and high-resolution mass spectrometry (HRMS) to characterize known and unknown per- and polyfluoroalkylated substances (PFAS) in foodstuffs	Nicolas Macorps	Oniris, INRAE, LABERCA	Calla 1

Wednesday 22 November

Time	Session	Title	Presenter	Organisation	Room
16:00-16:15		A strategy to evaluate food safety in circular food production systems	Milou G.M. van de Schans	Wageningen University and Research	Calla 1
16:15-16:30		Occurrence of high-volume production chemicals in highly consumed seafood species and evaluation of dietary intake and risk characterization	Sílvia Borrull	Universitat Rovira i Virgili	Calla 1
16:30-16:45		Polychlorinated alkanes (PCAs): Investigating levels in food from the Dutch market	Waut Bergkamp	Wageningen University and Research	Calla 1
16:45-17:00		Food analysis: towards greener and whiter analytical methods for the determination of inorganic contaminants	Mariela Pistón	Faculty of Chemistry, Universidad de la República	Calla 1
17:00-17:15		Pyrrolizidine alkaloids: Plant defence metabolites of serious health concern. An update on recent developments.	Patrick Mulder	Wageningen University and Research	Calla 1
	Parallel session - Micro/nano plastics analysis (Chairs: Marja Lamoree & Maria Margalef)				Calla 2
15:30-15:45		Chemometrics for improved identification and quantification of polymers in blood using Pyrolysis-GC-MS	Wilco Nijenhuis	Vrije Universiteit Amsterdam	Calla 2
15:45-16:00		Analysis of Micro- And Nanoplastic in Human Blood	Marthinus Brits	Vrije Universiteit Amsterdam	Calla 2
16:00-16:15		Towards simultaneous size determination and polymer identification of nanoplastics	Maria Hayder	University of Amsterdam	Calla 2
16:15-16:30		Towards FAIR data in the field of environmental research on microplastics	Montserrat Filella	University pf Geneva	Calla 2
16:30-16:45		Microplastics in urban freshwaters: a case study in the city of Amsterdam	Feride Öykü Sefiloglu	Vrije Universiteit Amsterdam	Calla 2
17:15-18:00	**Exhibition & posters**				Exhibition room
18:00-19:00	**General assembly IAEAC [members only]**				Calla 1

Thursday 23 November

Time	Session	Title	Presenter	Organisation	Room
	Keynotes (Chairs: Marja Lamoree & Laura Righetti)				
9:00-9:30		Advancing Green and Sustainable Analytical Chemistry for Environmental and Food Analysis	Charlotta Turner	Lund University	Plenary
9:30-10:00		Strategies for characterizing the consumer's chemical exposome	Gaud Dervilly	Oniris, INRAE, LABERCA, Oniris/INRAE	Plenary
10:00-10:15	Keynote Q&A				Plenary

Thursday 23 November

Time	Session	Title	Presenter	Organisation	Room
10:15-11:00	**Exhibition, Posters & Coffee**				Exhibition room
	Plenary session - Effect directed analysis and in vitro assays for mixture assessment (Chairs: Sam Nugen & Frederic Béen)				
11:00-11:15		Effect-directed analysis in the environment-food-human continuum to identify chemicals with Transthyretin binding properties	Maria Margalef Jornet	Vrije Universiteit Amsterdam	Plenary
11:15-11:30		Pull-down approach for the identification of endocrine-disrupting compounds in complex mixtures	Klara Hilscherova	Masaryk University	Plenary
11:30-11:45		The contribution of PFAS to thyroid hormone-displacing activity in Dutch waters: A comparison between two in vitro bioassays with chemical analysis.	Jaimy de Schepper	Het Waterlaboratorium, NL	Plenary
11:45-12:00		Optimizing analytical approaches in high-throughput effect-directed analysis (HT-EDA) for improved toxicity driver identification	Iker Alvarez-Mora	Helmholtz Center for Environmental Research (UFZ)	Plenary
12:00-12:15		Effect-directed analysis for multiple endpoints of bioactive compounds in the (drinking) water cycle	Corine Houtman	Het Waterlaboratorium, NL	Plenary
12:15-12:30		Novel Effect-Direct Analysis (EDA) approach to assess endocrine activity of food mixtures	Maricel Marin-Kuan	Nestlé Research, Societé des Produits Nestlé SA	Plenary
12:30-13:30	**Lunch in restaurant**				Restaurant
	Plenary session - Keynote & more (Chairs: Laura Righetti & Maria Margalef)				
13:30-14:00	Keynote	Endocrine disruptors in freshwater: policy perspectives by the OECD	Marijn Korndewal	OECD	Plenary
14:00-14:15	Analytics and sampling	Demonstrating the reliability of metabolomics-based chemical grouping: Towards acceptable practice	Pim Leonards	Vrije Universiteit Amsterdam	Plenary
14:15-14:30	Analytics and sampling	GC×GC for the determination of fatty acids in cow colostrum during the first days of lactation	Veronika Farková	Masaryk University, CZ	Plenary
14:30-14:45	Analytics and sampling	New sampling technique for determination and identification of microplastics in agricultural compartments and on its products with chemical imaging by FTIR- and Raman microscopy	Jürgen Schram	Niederrhein University of Applied Sciences	Plenary
14:45-15:30	**Exhibition, Posters & Coffee**				Exhibition room
	Parallel session - Food contact materials (Chairs: Gaud Dervilly & Hans-Gerd Janssen)				Calla 1
15:30-15:45		Human dietary exposure to food packaging contaminants through in vitro bioaccessibility studies	Letricia Barbosa-Pereira	University of Santiago de Compostela	Calla 1
15:45-16:00		Identification of non-intentionally added substances migrating from inner coatings of metallic cans – Toward provisional risk assessment of oligoesters	Ronan Cariou	Oniris, INRAE, LABERCA	Calla 1
16:00-16:15		Non-target screening of chemicals migrating from reusable sports plastic bottles into drinking water	Selina Tisler	University of Copenhagen	Calla 1

Thursday 23 November

Time	Session	Title	Presenter	Organisation	Room
16:15-16:30		UPLC-IMS-QTOF and SPME-GC-MS to quantify migrant species in vacuum cooking bags.	Carlos Jiménez Estremera	University of Zaragoza	Calla 1
16:30-16:45		Strategy for Ensuring the Safety of Novel Packaging Materials	Elsa Omer	Nestlé Research, Societé des Produits Nestlé SA	Calla 1
16:45-17:00		Evaluation of new approaches to decontaminate recycled polyolefins destined to come into contact with food products	Margarita Aznar	University of Zaragoza	Calla 1
17:00-17:15		Combined untargeted screenings of substances migrating from emerging food contact materials	Salvatore Ciano	Sciensano	Calla 1
	Parallel session - Toxic chemicals, metabolites and transformation products (2) (Chairs: Timur Baygildiev & Maria Margalef)				Calla 2
15:30-15:45		Unravelling the early exposure to PFAS in children at ages 4 and 8 from INMA Gipuzkoa cohort.	Anne San Roman	IIS Biodonostia	Calla 2
15:45-16:00		Trace element speciation made easy: introducing Frontal Chromatography ICP-MS	Damiano Monticelli	Università degli Studi dell'Insubria	Calla 2
16:00-16:15		Thermal processes and secondary recycling regulate the atmospheric levels of highly toxic polychlorinated naphthalenes in an urban Mediterranean site	Minas Iakovides	The Cyprus Institute	Calla 2
16:15-16:30		Passive sampling of high production volume chemicals and polycyclic aromatic hidrocarbons in outdoor air samples. Application and risk assessment evaluation	Reyes Garcia	Universitat Rovira i Virgili	Calla 2
16:30-16:45		Development of a novel passive sampler based on membrane assisted solvent extraction and molecularly imprinted polymer for monitoring of selected pharmaceuticals in surface water	Sinegugu Khulu	University of the Witwatersrand	Calla 2
16:45-17:00		Investigating the sorption capacity of four Nigerian clays for the removal of arsenic, cadmium, chromium, and lead from water.	Augustine Imogben Umarein	University of Strathclyde, Glasgow	Calla 2
17:00-17:15		Determination of haloacetic acids in reclaimed water and drinking water by direct injection and hydrophilic interaction chromatography coupled with mass spectrometry	Eva Hernández Jambrina	Joint Centre University of Almeria-CIEMAT	Calla 2
17:15-18:00	**Exhibition & posters**				Exhibition room
18:30-21:30	**Conference dinner**				

Friday 24 November

Time	Session	Title	Presenter	Organisation	Room
	Keynotes (Chairs: Maria Margalef & Hans Mol)				
8:45-9:15	Advances on the identification, monitoring, and prioritization of persistent, mobile and toxic (PMT) substances threatening water resources		Hans-Peter Arp	NTNU	Plenary
9:15-9:45	New Challenges and Opportunities in Food Analysis		Katerina Mastovska	AOAC international	Plenary
9:45-10:15	Towards sample-to-solution approaches for food contaminant determination at the point-of-need (Pittcon contribution)		Gert Salentijn	Wageningen University and Research	Plenary
10:15-10:30	Keynote Q&A				Plenary
10:30-11:00	**Exhibition, Posters & Coffee**				Exhibition room
	Plenary session - Sampling strategies and on-site detection (Chairs: Montserra Filella & Hans Mol)				Plenary
11:00-11:15	Combatting cannibalism: early detection of processed animal proteins in poultry feed		Marleen Voorhuijzen	Wageningen University and Research	Plenary
11:15-11:30	Three sampling strategies for microplastics analysis in beaches: A critical comparison		Thomas Maupas	University of the Basque Country	Plenary
11:30-11:45	Towards rapid on-site detection of atropine in cereals		Ids Benjamin Lemmink	Wageningen University and Research	Plenary
11:45-12:00	Experimental and computational optimization of polymer inclusion membrane technique for mineral extraction in seawater		Luke Chimuka	University of the Witwatersrand	Plenary
12:00-12:45	**Closing ceremony: announcement ISEAC-42, best oral and poster awards**				Plenary