Standard ISO/CEN methods for monitoring microplastics

Example of analysis of microplastics in drinking water by vibrational spectroscopies for rationalizing the debate through ISO normalization

NIZAR BENISMAIL

NESTLE WATERS

EUROQCHARM FINAL CONFERENCE - OCT 2023

AGENDA

1 Context of MicroPlastics

2 Methodologies & challenges for MPs analysis

3 Water method harmonization as opportunity for first standard

Microplastics interest

Google research with “Microplastics”

Microplastics interest

WHO report on microplastics in drinking water

Tiny microplastic particles can be absorbed through plant roots and end up in the wheat and vegetables we eat, study shows

- Researchers studied ways microplastics are able to enter the human food chain
- They found that some particles were entering the roots of human edible food
- Plastic pieces reached the plants after accumulating in soil and wastewater

CONSCIOUS CONSUMER

Microplastics in Food, Water & Air: Can We Avoid Them?

By Sarah Mosko January 24, 2020

You're likely taking in tiny particles of plastic according to research into the risks to human the environment. Although scientists have yet reason enough to worry.

Microplastics (MP) result from the breakdown into ever smaller bits of everyday plastic discards, like packaging, children's toys, and synthetic clothing and carpeting. Despite their small dimension (sometimes invisible), MP are still made of long-chain polymer molecules that make plastics resistant to bio-degradation.

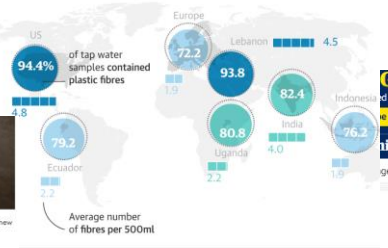
News > Environment

Boiling a litre of water in a plastic kettle releases 10m microplastics into our tea and coffee, study finds

35% of microplastics in oceans come from clothing, research reveals

A new report by the Institution of Mechanical Engineers has revealed that 35% of microplastics released into the world's oceans are from synthetic textiles.

Tap water is widely contaminated by plastic



FILTERING

How to filter and remove microplastics from tap wate

Salt is produced on Madura Island, Indonesia, by evaporating seawater, an ancient technique. A new study found that salt made in this region contains some of the highest microplastics sampled.

PHOTOGRAPH BY ULET IFANSASTI, GETTY IMAGES

ENVIRONMENT | PLANET OR PLASTIC?

Microplastics found in 90 percent of table salt

A new study looked at sea, rock, and lake salt sold around the world. Here's what you need to know.

Guardian

Search jobs

Sport Culture Lifestyle

Wildlife Energy Pollution

Bottle-fed babies swallow millions of microplastics a day, study finds

Exposure is far higher than previously though plastic food containers

Home > Topics > Lifestyle Monit

Those fancy tea bags? Microplastics in them are macro offenders

New study finds nylon tea bags leech billions of microplastics into every single cup of tea

The New York Times

Opinion

You're Probably Inhaling Microplastics Right Now

A new study found plentiful evidence of these tiny particles in dust in the nation's most remote places.

By Janice Brahney

Dr. Brahney is a biogeochemist at Utah State University.

June 25, 2020

Microplastics are everywhere, study finc

by Birgitte Svennevig, University of Southern Denmark

How Synthetic Microfibers End Up In Food and Drinking Water

SPONSORED CONTENT

By Catherine Saffino

Vehicle tyre particles 'significant' source of marine microplastic pollution

01.06.2020

NEWS

Not only do they contribute to air pollution in our towns and cities, particles released from vehicle tyres could also be a 'significant' source of microplastics in the world's oceans, a ground-breaking study has suggested.

Credit: CC0 Public Domai

Microplastics are eve air that we breathe. H countries, researchers are worried.

LE FIL VERT

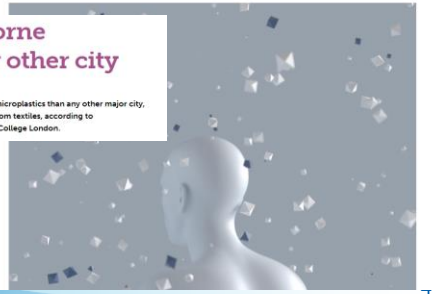
«Il y a dix fois plus de microplastique dans l'air intérieur qu'extérieur»

London has more airborne microplastics than any other city

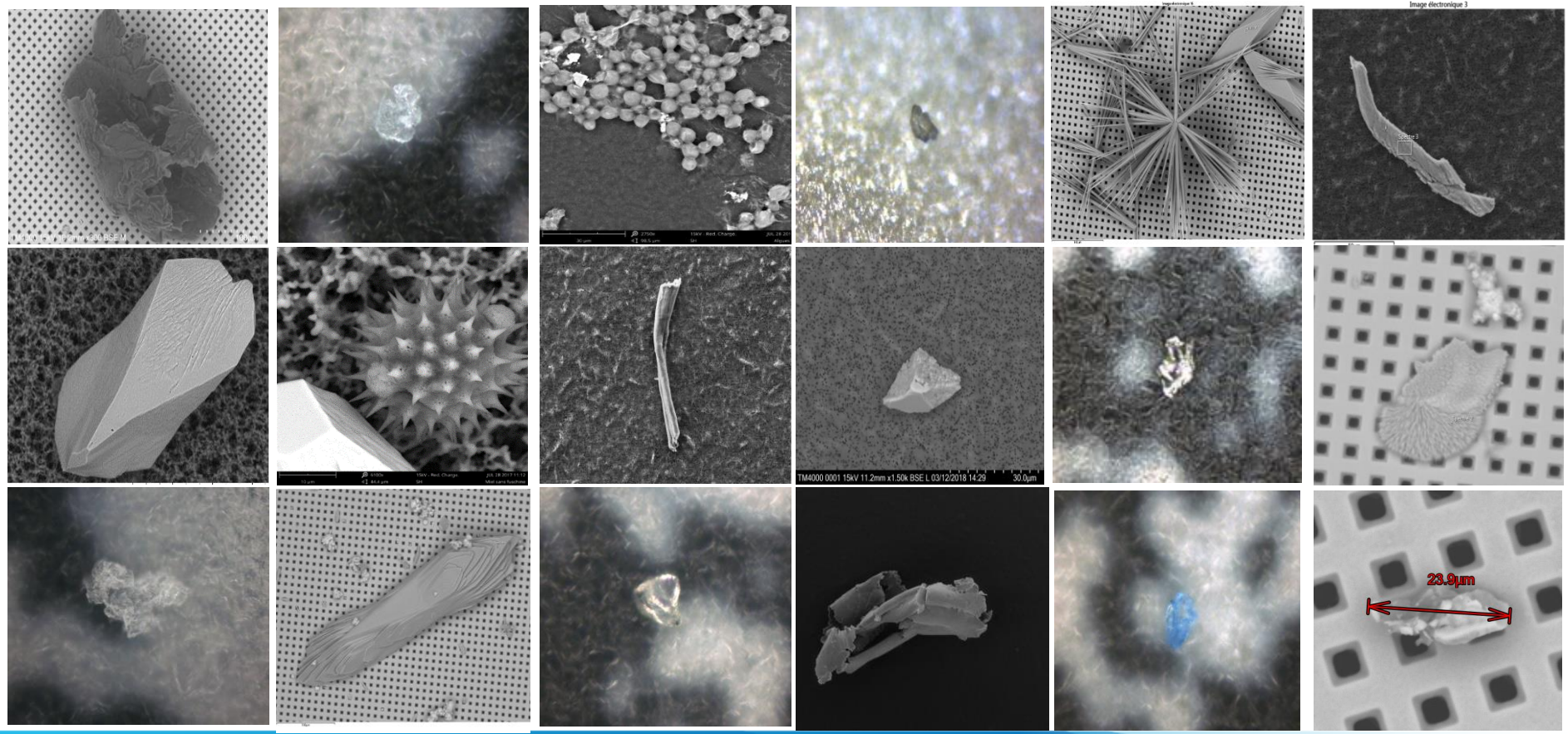
05.01.2020

HEALTH

London has the highest level of airborne microplastics than any other major city, with 92% of these microplastics coming from textiles, according to a study published by researchers at Kings College London.



Are these particles / UFOs microplastics ?



1 Context of MicroPlastics

2 Methodologies & challenges for MPs analysis

3 Water method harmonization as opportunity for first standard

Microplastics methods : where were we ?

At the beginning

First studies used a microscope : all that seemed plastic to be “this is plastic”

Slowly better methods evolved but not yet standardized methods

Much of the published data is dubious : many scientists say “we can measure it” but few feel the need to prove that they measure correctly

May 2019

Microplastics in Freshwaters and Drinking Water:

Critical Review and Assessment of Data Quality

Albert A. Koelmans[†,*], Nur Hazimah Mohamed Nor[†], Enya Hermsen[†], Merel Kooi[†], Svenja M. Mintenig[‡,§], Jennifer De France[‡,*]

[†]Aquatic Ecology and Water Quality Management Group, Wageningen University, The Netherlands.
[‡]Copernicus Institute of Sustainable Development, Utrecht University, The Netherlands.
[§]KWR Watercycle Research Institute, Nieuwegein, The Netherlands.
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3.3 Conclusions

We conclude that based on the limited number of high quality studies identified, standardization of microplastic analysis in water is needed. Quality assurance criteria that require the most improvements are sample treatment, polymer identification, laboratory preparation, clean air conditions and positive controls. In addition to ensuring that individual studies are of higher quality in order to achieve more confidence in study findings, standardized methods will allow reproducibility and comparability of results and will lead to the quality of data that are needed to conduct risk assessments. Among water types,

Microplastics – Technical Working Group on clean water – June 2018

Chemical and Veterinary Analytical Institute Muensterland-Emscher-Lippe

idaea CSIC

Technische Universität München

Bavarian Health and Food Safety Authority

European Commission

TZW Technologiezentrum Wasser

Ifremer

ipf Leibniz-Institut für Polymerforschung Dresden

DANONE WATERS

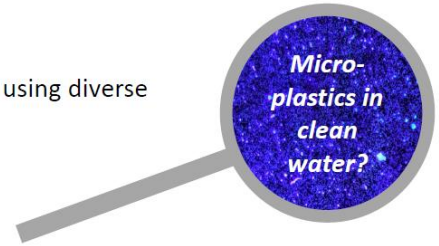
SGS INSTITUT FRESENIUS

Nestlé Quality Assurance Center

Vittel

WWU MÜNSTER

Abstract/Intention



Lots of studies on microplastics in water are published using diverse analytical techniques.

→ The results are not comparable or even invalid.

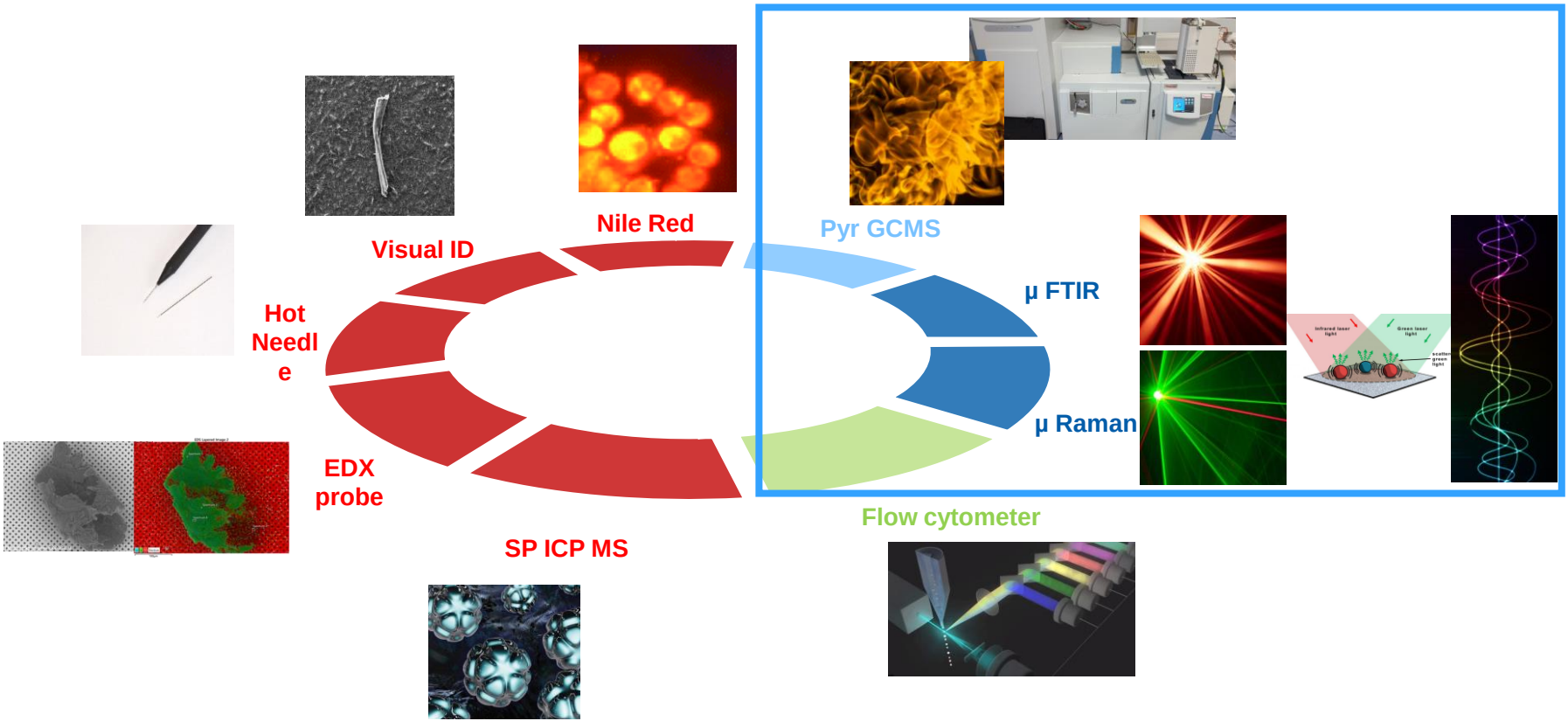
→ There is a huge need for harmonization.

We established a working group of 12 laboratories experienced in microplastic analysis.

Minimum requirements and **best practices** were discussed and specified as consensus for microplastic analysis in clean water (low matrix content).

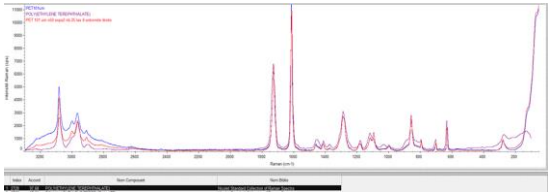
Poster & Scientific publication
B.Ossmann, D. Schymanski & al.

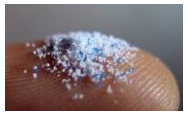
Microplastics methods : existing methodologies in scientific journals/media



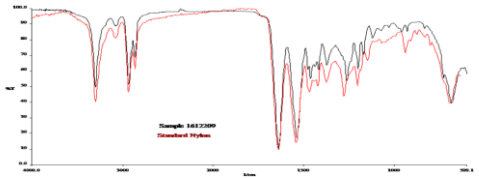
Microplastics methods : adequate equipments

Micro-Raman Microscopy : Organic, polymeric and cristallin structure



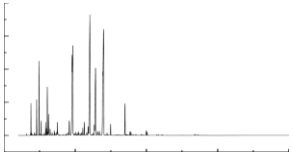


Micro-FTIR Microscopy : Organic, polymeric and cristallin structure



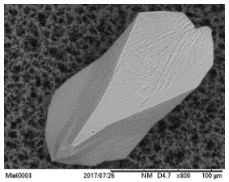
In case of high amout of MPs in complex environmental matrix

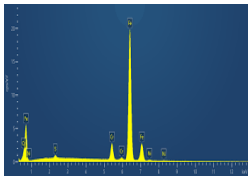
Pyr GC-MS : organic structure and quantification



Complementary identification

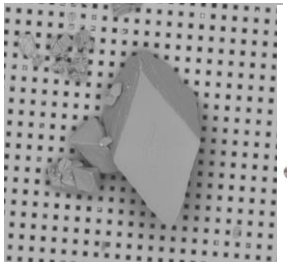
MEB-EDS : electronic microscopy and EDX probe for elemental composition





Microplastics methods : technical summary of where we stand

- Although microplastics (MP) are recognized as an emerging contaminant in the environment
  - No standardized analytical methods : currently neither sampling, extraction, purification nor identification approach are standardized, making the increasing numbers of MP studies hardly comparable
  - No Regulatory limit for drinking water
- Only reliable identification but time-consuming methods through Pyr-GC/MS or Spectroscopy (Micro-Raman or Micro-FTIR)
- As microplastics ubiquitous in environment (Air), difficult to have lab blank/reference non polluted

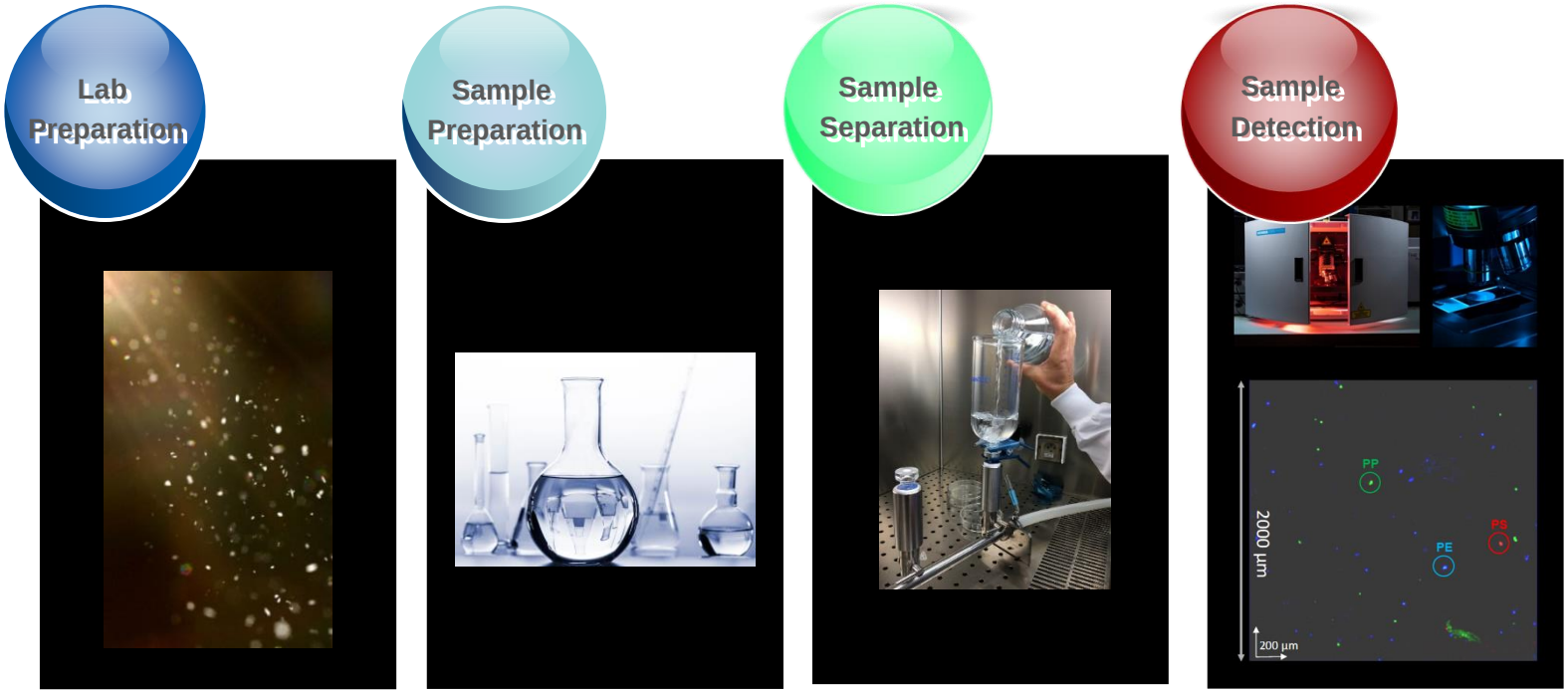


Let's take the example of the easiest matrix to analyze : clean water

We want : reliable, fast and reproducible microplastic quantification

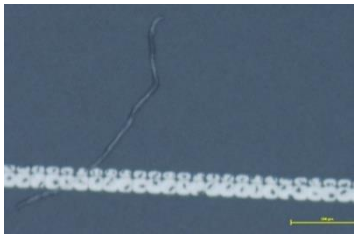
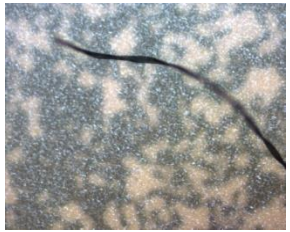
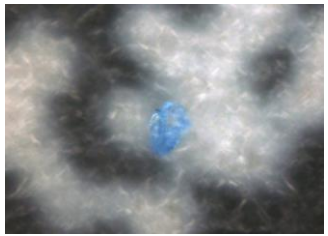
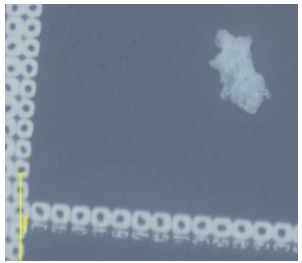
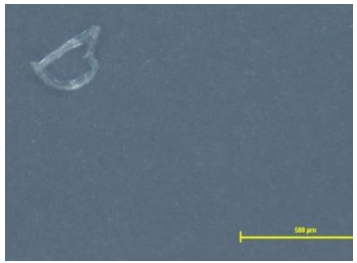
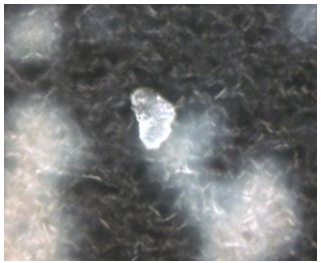
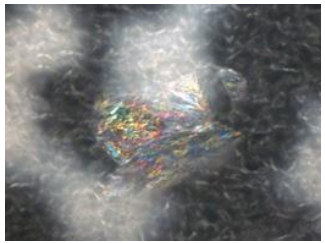
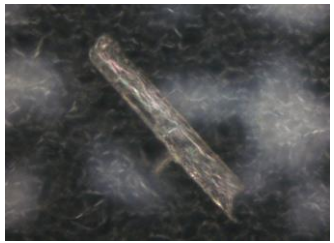
- Improved sample preparation – faster, better recovery, low contamination
- Improved full automated microplastic detection
- Results as number & polymer identity at particle size down to 1 to 10 µm

Microplastics spectroscopic methods : extremely challenging to develop



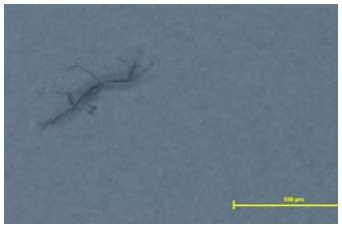
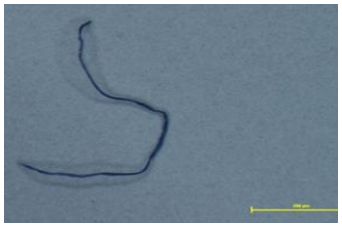
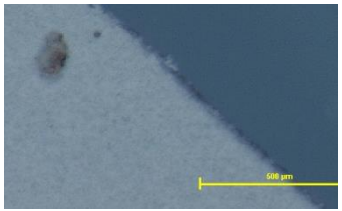
Developing reliable method to count and identify MPs is extremely challenging

Microplastics methods : examples of cross contamination from labs

Blank reference

Air

Microplastics methods : fallpits in blank contamination

Controled atmosphere

No plastic mask wearing

No use of make-up, cream,vernish, toothpaste or shampoon containing MPs

Hand wash and no gloves

Cotton labcoat – avoid synthetic clothes

Cleaned glass or inox system – no plastic consumable or part touching the sample

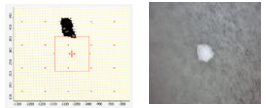
All products used for preparation step must comply with purity or be filtred

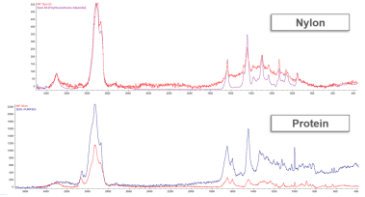
External cleaning of the sample

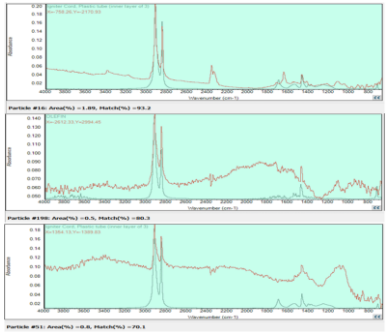
Protection of sample during transportation & analysis

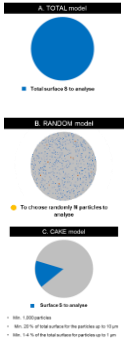
Microplastics spectroscopic methods : fallpits in signal acquisition

destruction and black carbon creation

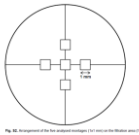








Statistically representative ?



% of filter surface analyzed

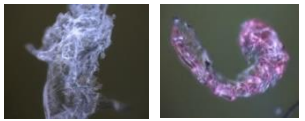
Overpower of laser

Vibrations & focus issues

False positives

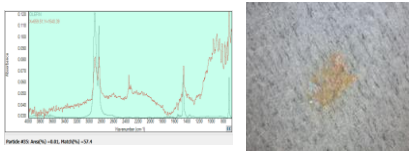
Identification matching from algorithm

Pigment crossed signal



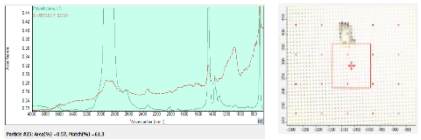
Particle thickness

High fluorescence

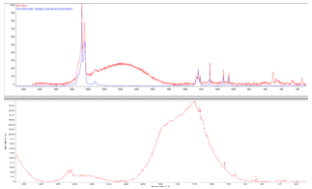


Low caracteristic signal

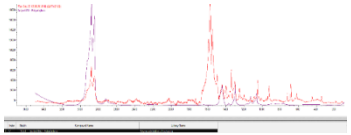
Contrast Brightnes



Generic adjustment needed



hump of fluorescence



Microplastics spectroscopic methods : data analysis optimization

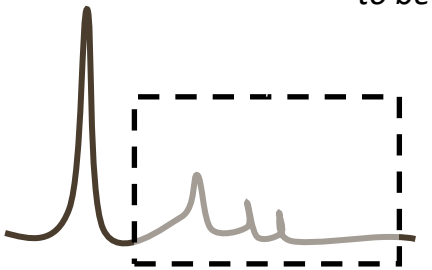
- ✓ *Background substruction*
- ✓ *Atmospheric correction*
- ✓ *Baseline correction*
- ✓ *Smoothing*
- ✓ *…*

- ✓ *Pearson correlation*
- ✓ *Euclidean distance*
- ✓ *Linear of quandratic discriminant analysis*
- ✓ *Machine learning*

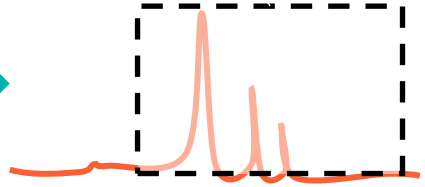
TO BE ADAPTED BY LABORATORY

Microplastics spectroscopic methods : interferences in signal acquisition

Only informative part of spectra to be considered



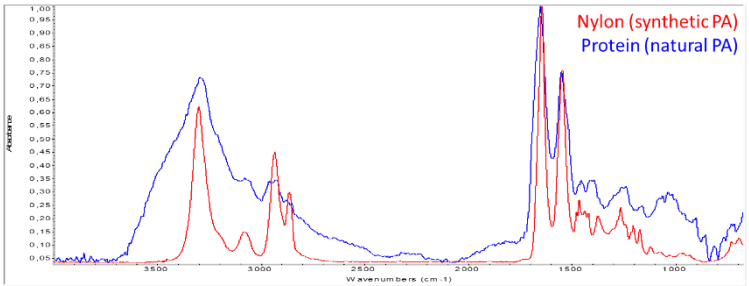
Processed spectrum



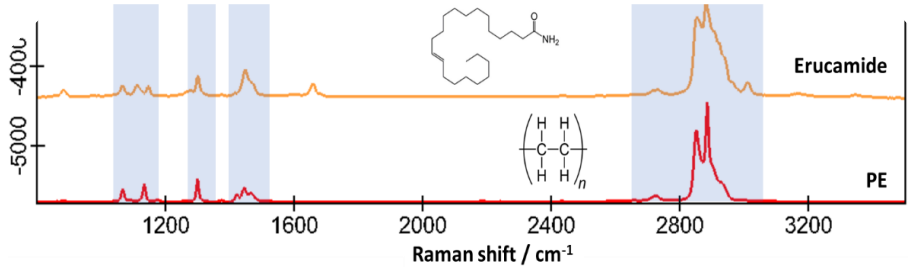
Database

- ✓ *Same acquisition mode as the raw spectra*
- ✓ *Polymers with additives, aged…*
- ✓ ***Other organic and inorganic compounds***

Interference: Nylon vs Proteins



Interference: PE vs fatty acids/amides



Microplastics spectroscopic methods : all steps optimized under TWG

Sampling
- Type of container, Volume sample, replication
- Preparation of container, specific precaution

Treatment
- Lab working environment (Air, hood, lab coat, consumables, tools, equipment, cleaning of external part…)
- Addition of reagent, homogenization

Filtration
- Filtration system, nature of filter, coating, dimension , filtration steps, filter holder, cleaning/rinsing protocol, volume of filtration, handling and transport of filter, storage of the filter

Analyse by Raman/IR
- Hardware, software, particle detection mode, objective, focus, contrast, light,
- IR acquisition mode, acquisition range, wavelength, beam spot, laser power, repetition, spectral resolution, scan number, background correction, fluorescence correction
- Surface of filter analysed, number of zone, area geometric

Results / Report
- Database libraries, own database, match acceptance criterion
- Frequency of blank analyse, blank expression in results (blank average, blank subtraction…), limit of detection/quantification
- Type, number, size, distribution of Microparticles and Microplastics, shape, colour, other particles (proteins, minerals, cellulosic, unknowns…)

Validation/ p-tests
- Limit of detection/quantification, blank management, repeatability, reproducibility, specificity, accuracy, recovery
- False positives, false negatives
- Proficiency tests

Minimum Requirements And Best Practices

ANALYTICAL & BIOANALYTICAL CHEMISTRY

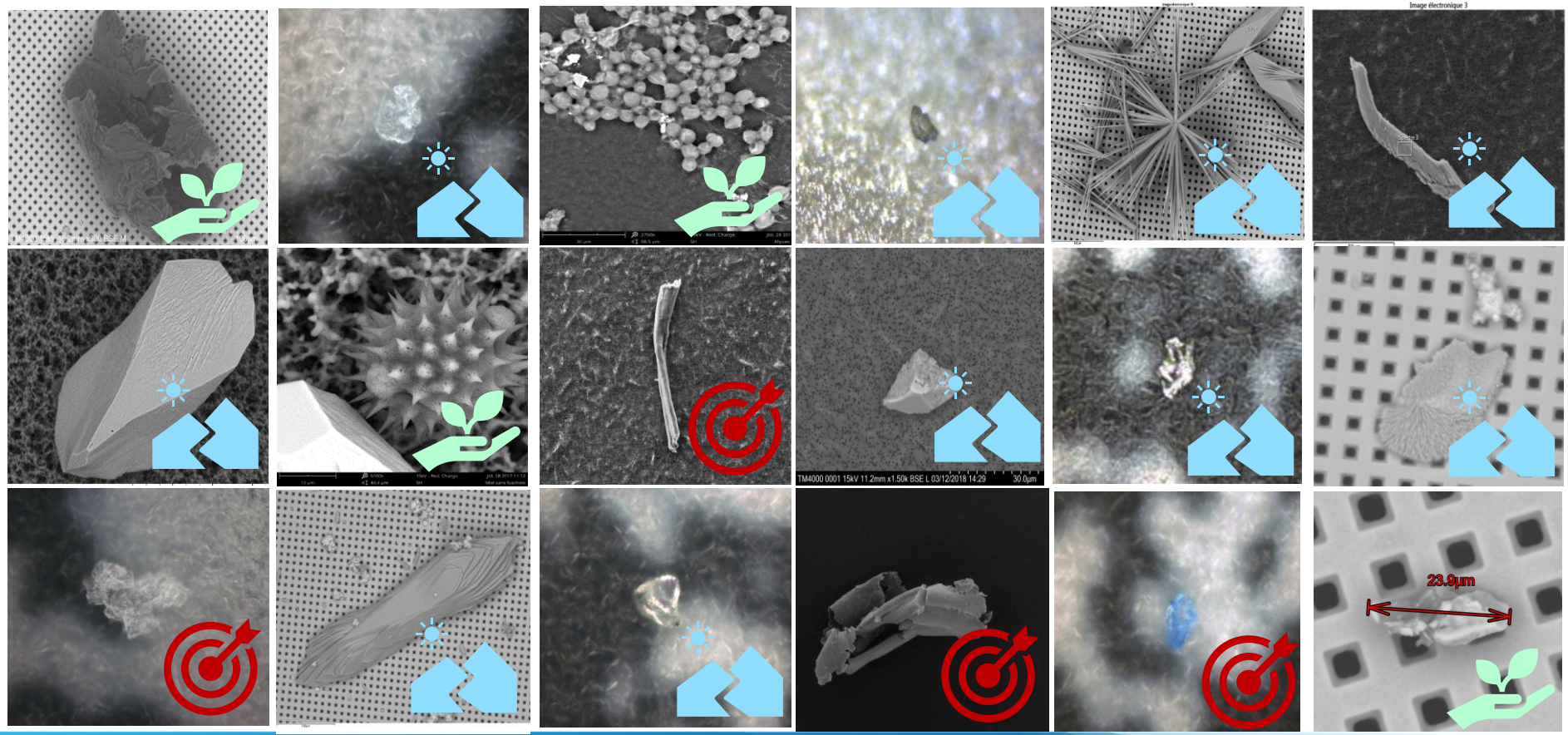
Springer

Analysis of microplastics in drinking water and other clean water samples with micro-Raman and micro-infrared spectroscopy: Minimum requirements and best practice guidelines

Journal:	*Analytical and Bioanalytical Chemistry*
Manuscript ID	ABC-00671-2021.R1
Type of Paper:	Critical Review
ate Submitted by the Author:	n/a
plete List of Authors:	Schymanski, Darena; Chemical and Veterinary Analytical Institute Münsterland-Emscher-Lippe (CVUA-MEL) Oßmann, Barbara; Bayerisches Landesamt fur Gesundheit und Lebensmittelsicherheit, Benismail, Nizar; Nestle Quality Assurance Center Vittel Dallmann, Gerald; SGS Institut Fresenius GmbH von der Esch, Elisabeth; Technical University of Munich, Institute of Hydrochemistry, Chair for Analytical Chemistry and Water Chemistry Fischer, Dieter; Leibniz Institute of Polymer Research Dresden, Fischer, Franziska; Leibniz Institute of Polymer Research Dresden (IPF) Gilliland, Douglas; European Commission Joint Research Centre Ispra Sector Glas, Karl; Chair of Food Chemistry and Molecular Sensory Science, Technical University of Munich Käppler, Andrea; SGS Institut Fresenius GmbH Lacorte, Silvia; Institute of Environmental Assessment and Water Research, Department of Environmental Chemistry Weisser, Jana; Chair of Food Chemistry and Molecular Sensory Science Witzig, Cordula; TZW: DVGW-Technologiezentrum Wasser (German Water Centre) Zumbülte, Nicole; TZW: DVGW-Technologiezentrum Wasser (German Water Centre), Ivleva, Natalia; Technische Universität München, Institute of Hydrochemistry, Chair for Analytical Chemistry and Water Chemistry;
Keywords:	microplastic, micro-Raman spectroscopy, micro-(FT)IR spectroscopy, bottled water, drinking water, clean water

https://doi.org/10.1007/s00216-021-03498-y
https://link.springer.com/content/pdf/10.1007/s00216-021-03498-y.pdf.

Are these particles / UFOs microplastics ?

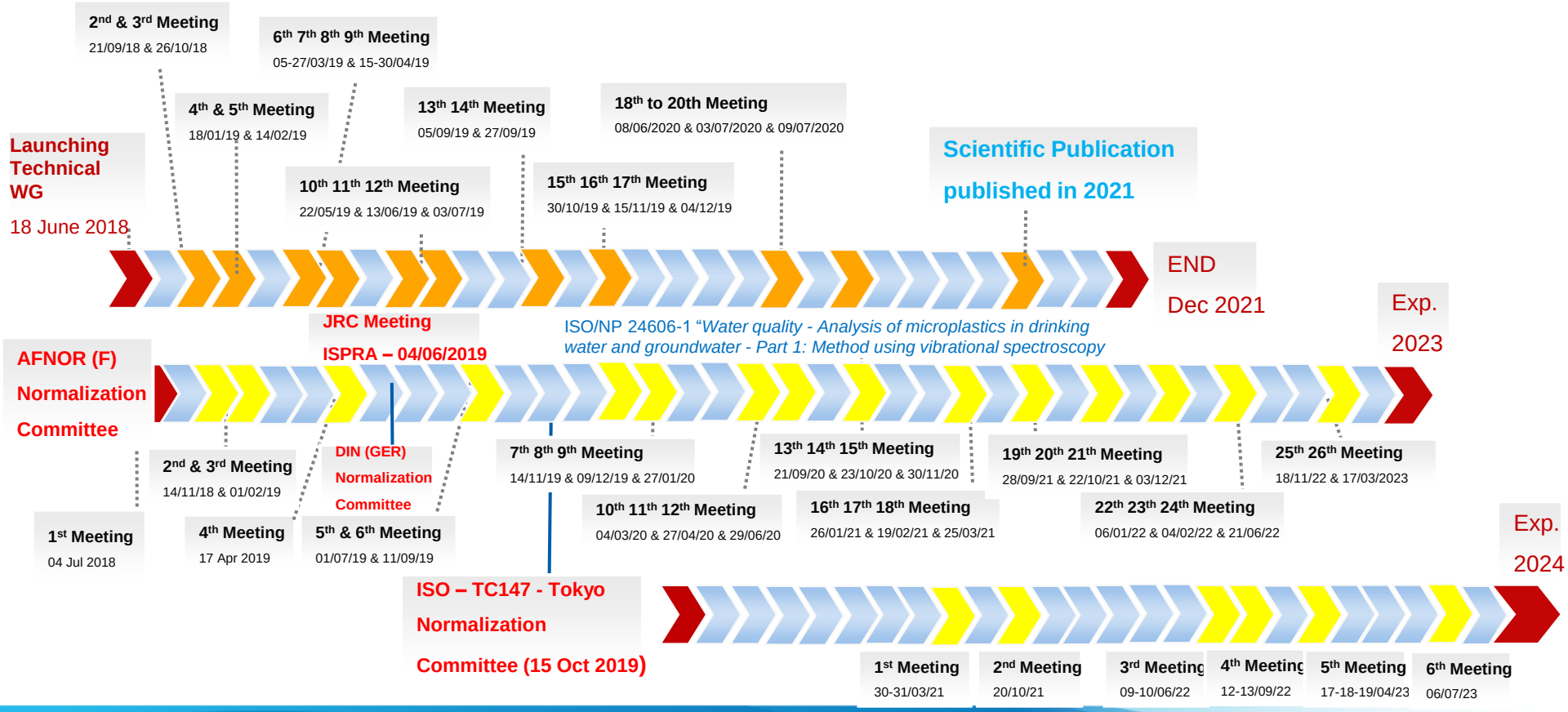


1 Context of MicroPlastics

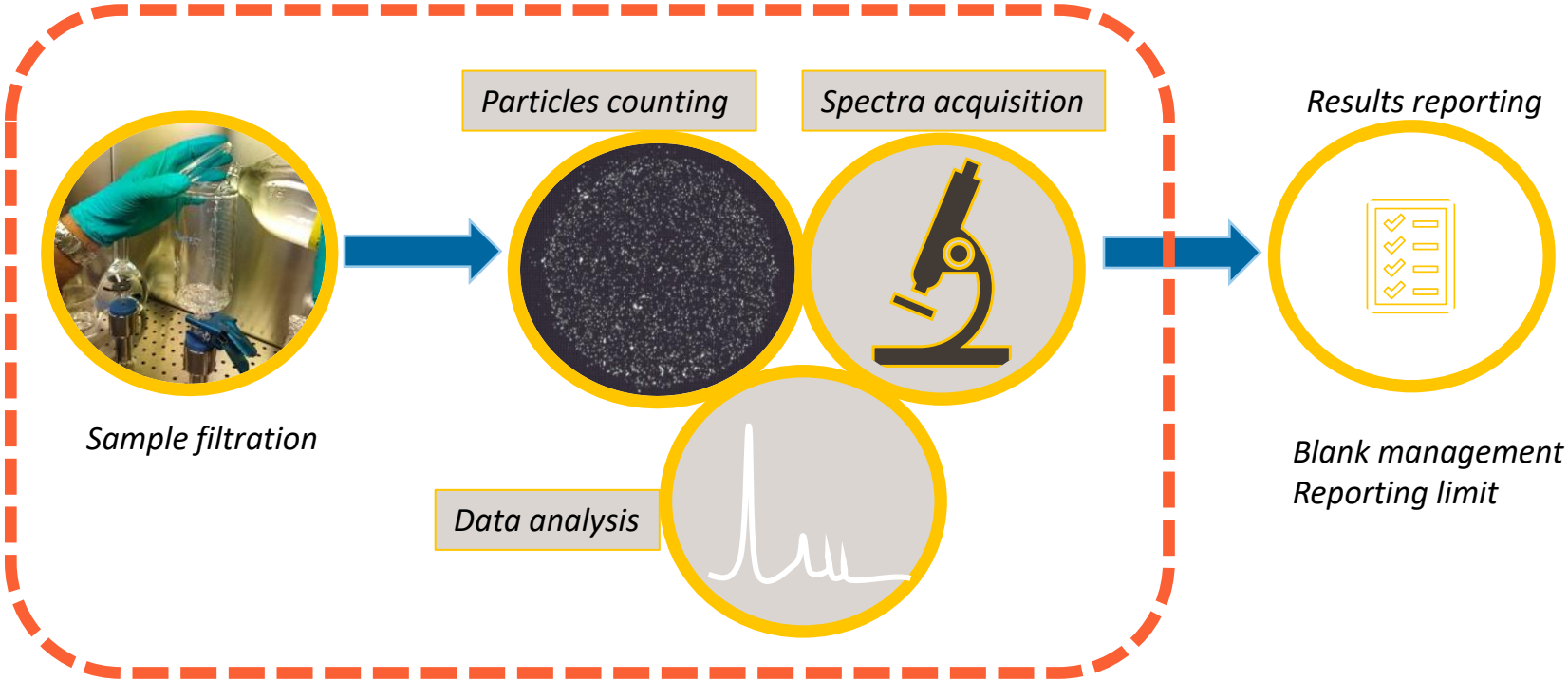
2 Methodologies & challenges for MPs analysis

3 Water method harmonization as opportunity for first standard

Microplastics - from TGW to ISO Normalization : a long journey

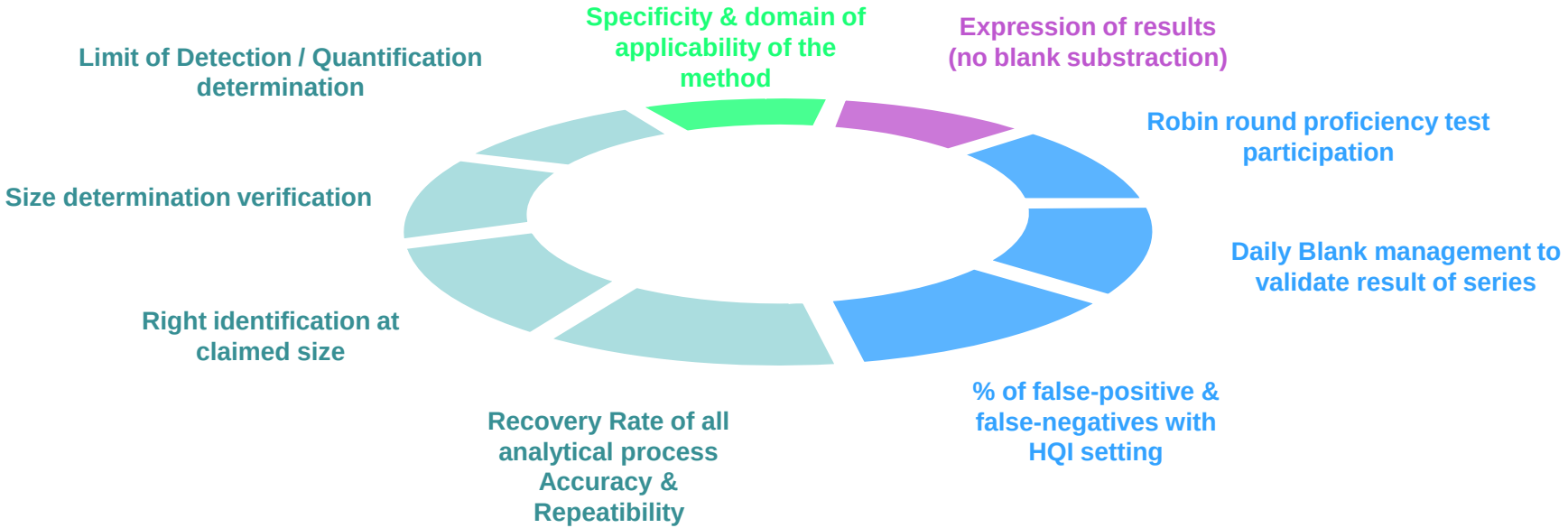


Microplastics - from spectroscopic methods to ISO standard



METHOD VALIDATION

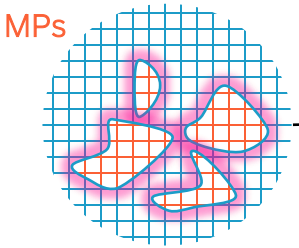
Microplastics « Spec » standard : validation & verification of the method



Microplastics « Spec » standard : verification of particle size measurement

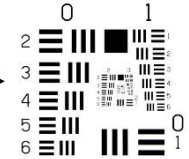
Type of image to be processed:

Optical Image

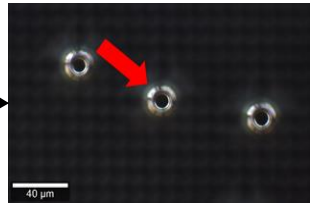


Spectral mapping

Examples of certified standards:



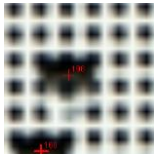
resolution test chart

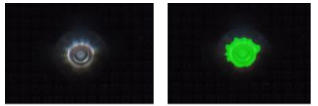


monodisperse beads

What affects the size measurement?

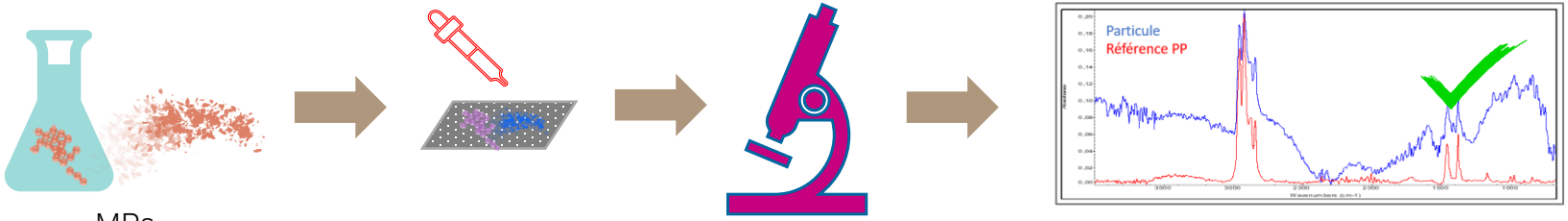
- Image resolution (pixel size)
- Contrast (or spectral)
- treshhold
- Filter pattern dimensions
- Particle artefacts
- Particles agglomeration





Define the trueness of the size measurement

Microplastics « Spec » standard : verification of right identification at claimed size



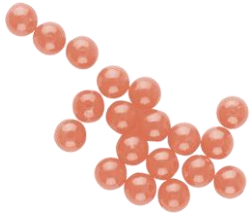
MPs standards

Optimized parameters of spectra acquisition

Optimized algorithm and HQI

… the laboratory must record the spectrum and correctly identify at least 4 different types of microplastics at the claimed size (lower limit of size)

Microplastics « Spec » standard : verification of recovery rate at claimed size



Plastic beads:
known quantity (N)
close to the claimed size

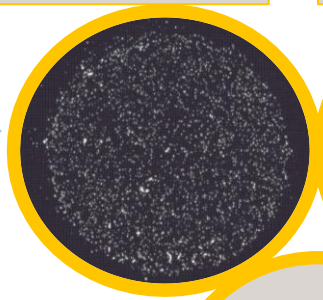
Recovery rate = $N_{\text{after filtration}}$ / N · 100%

RECOVERY RATE > 70 %

Microplastics « Spec » standard : results report

Sample filtration

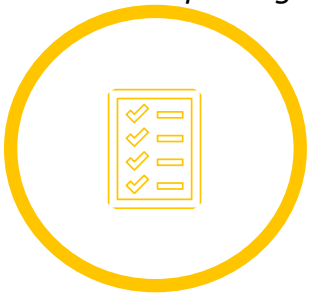
Particles counting



Spectra acquisition

Data analysis

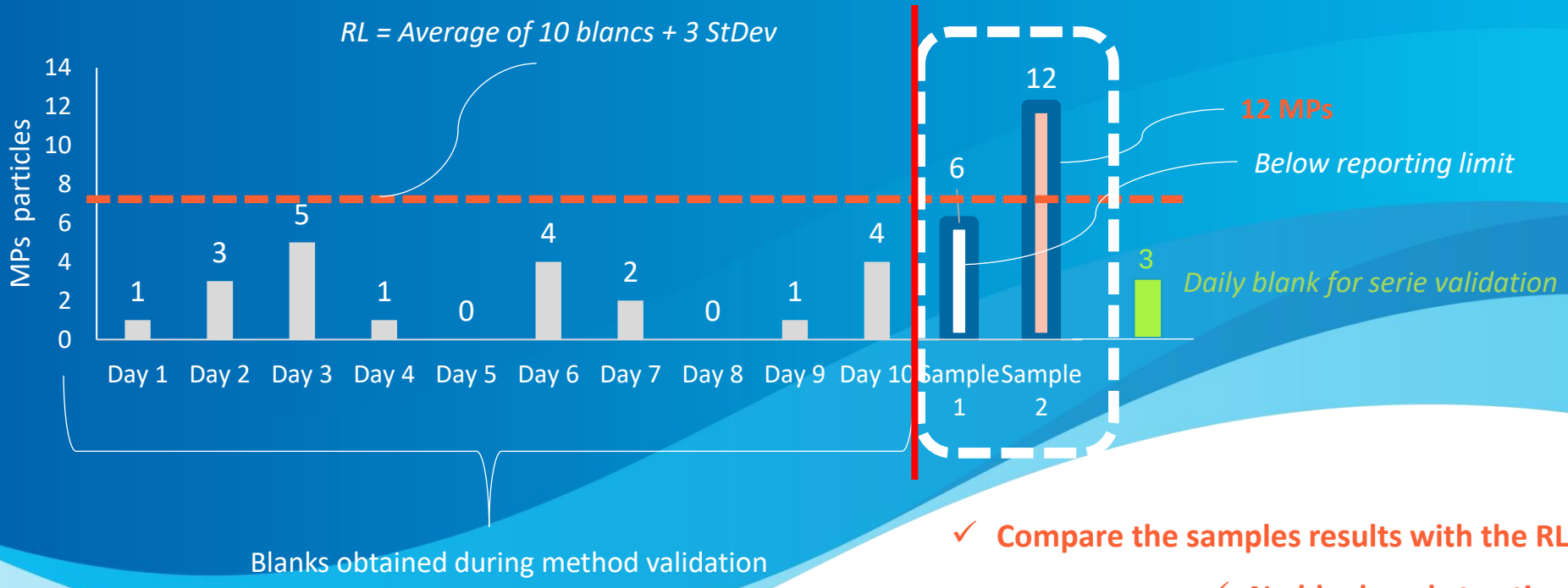
Results reporting



Blank management
Reporting limit

QUALITY CONTROL

Microplastics « Spec » standard : how to define and use reporting limits



✓ **Compare the samples results with the RL**

✓ **No blanks substraction**

✓ **If the daily blank of the serie > RL, invalidate the filtration serie**

Microplastic : detailed structure of norm ISO 16094-2 : 2023 (Spectroscopy)

Contents

- **ISO/DIS 16094-2:2023 —**

Water quality — Analysis of microplastic in water — Part 2: vibrational spectroscopy methods for waters with low content of suspended solids including drinking water

- analysis of microplastics by vibrational spectroscopy methods (Raman, µ-InfraRed instruments): generate information about particle size, particle type and particle numbers
- detection of microplastics in waters with low content of natural suspended solids, e.g. drinking water, groundwater or laboratory water.
- very specific technical description for harmonizing all the analytical steps: choices of filters, precautions to minimize cross contamination, calculation of the blank and limit of reporting of the laboratories, statistical models to deliver comparable results, data treatment, interpretation and reporting
- Inclusion of mandatory controls / validation procedures

Microplastics : ISO TC147 « Water Quality »

ISO/TC 147 – SC6 – WG 16

- ISO/CD 5667-27, Water quality – Sampling – Part 27: Sampling for microplastic particles and fibres in water
  => *Very general for different sorts of water, without specific recommendations*

ISO/TC 147 – SC2 – JWG 1 with ISO TC 61 (Plastics)

Title: Plastics (including microplastics) in waters and related matrices

Convenor: Nizar Benismail (F); Nestle Waters (previous Convenor: C.G. Bannick (D)) / Ulrike Braun (D); German Environment Agency

Countries: Austria, Belgium, Croatia, Finland, France, Germany, Italy, Japan, Korea, the Netherlands, Norway, Portugal, Spain, Sweden, Switzerland, United Kingdom, United States

Liaisons: European Commission, ECOS (Environment), UILI, VAMAS, IEC, ISO/TC 45 “Rubber and rubber products, ISO/TC 334 “Reference materials”, ISO/TC 146 “Air Quality”, ISO/TC 61/SC 14 “Plastics”

Scope: Standardization of methods for the characterization and quantification of plastics including microplastics and related polymers in water ...
NOTE: The JWG 1 offers to other interested technical committees to cooperate in the development and application of methods and methodologies elaborated in this group

- Series of documents ISO - 16094: 2023 Water quality — Analysis of microplastics

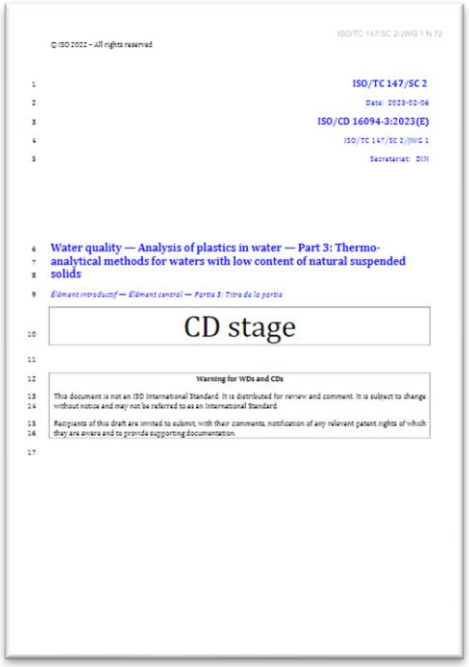
Microplastics : ISO TC147 « Water Quality » & CEN

- On going process for standardization at FR (AFNOR) and ISO Level now with ISO TC147 / SC2 / JWG1 :

- Series of ISO 16094 Standards under Vienna agreement : **Water quality - Analysis of microplastics**
  - Part 1: General ~~and sampling~~ (CD state)
  - Part 2: Vibrational spectroscopy (DIS state)
  - Part 3: Thermoanalytical methods (CD state)
  - Part 4: Sample preparation methods (On going)
  - Part 5: Ecotoxicological methods (To start)

ISO 16094 – Part 2 :
- By Sept 2023 DIS Submission (Enquiry)
- From Oct 2023 to December 2023 ; interlaboratory trial for norm validation
- By March 2024 final limit for Publication

Microplastics - Thermoanalytical methods ISO/CD 16094-3: 2023



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ISO/TC 147/SC 2

Date: 2023-02-06

ISO/CD 16094-3:2023(E)

ISO/TC 147/SC 2/WG 1

Secretariat: DIN

Water quality — Analysis of plastics in water — Part 3: Thermo-analytical methods for waters with low content of natural suspended solids

Élément introductif — Élément central — Partie 3: Titre de la partie

CD stage

Warning for WDs and CDs

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Recipients of this draft are invited to submit, with their comments, notification of any relevant patent rights of which they are aware and to provide supporting documentation.

- **ISO/CD 16094-3:2023 —**

Water quality — Analysis of microplastic in water — Part 3: thermo-analytical methods for waters with low content of suspended solids including drinking water

- general recommendations for the analysis of microplastics by thermo-analytical methods coupled with analysis of decomposition gases: determination of mass fractions and polymer type.
- detection of microplastics in waters with low/moderate content of natural suspended solids.
- general aspects for sample preparation and the application of thermoanalytical methods, and also includes related terms and definitions.
- very detailed technical description for the investigation of water filtrates using thermal extraction desorption gas chromatography/mass spectrometry (TED-GC/MS) and pyrolysis gas chromatography/mass spectrometry (Py-GC/MS) and investigation of isolated particles using Py-GC/MS.

Microplastics : Running standardization activities about “Microplastics”

	ISO		CEN
Area „plastics “	ISO/TC 61 “Plastics” ISO/TC 61/SC 14 “Environmental aspects” WG 4 Characterization of plastics leaked into the environment (including microplastics) and quality control criteria of respective methods	EN ISO 17422:2018 Plastics – Environmental aspects – General guidelines for their inclusion in standards CEN ISO/TR 21960: 2020 Plastics – Environmental aspects – State of knowledge and methodologies EN 17615:2022 Plastics – Environmental Aspects – Vocabulary ISO/FDIS 24187: 2023 Principles for plastic and microplastic analysis present in the environment	CEN/TC 249 Plastics WG 24 Environmental aspects
Area „water“	ISO/TC 147 Water Quality SC 2 “Physical, chemical and biochemical methods” JWG 1 Joint ISO/TC 147/SC 2 - ISO/TC 61/SC 14 WG: Plastics (including microplastics) in waters and related matrices SC 6 “Sampling (general methods)” WG 16 Sampling for microplastics	ISO/CD 16094-1 **ISO/DIS 16094-2:2023 — Water quality — Analysis of microplastic in water — Part 2: vibrational spectroscopy methods for waters with low content of suspended solids including drinking water** ISO/CD 16094-3 ISO/PWI 16094-4 **ISO/CD 5667-27, Water quality – Sampling – Part 27: Sampling for microplastic particles and fibres in water**	*Agrees to takeover documents of ISO under Vienna agreement*
Area „textiles“	ISO/TC 38 Textiles WG 34 Microplastics from textile sources WG 35 Environmental aspects	ISO/DIS 5157, 2022 Textiles – Environmental aspects – Vocabulary EN ISO 4484-1: 2023 Textiles and textile products – Microplastics from textile sources – Part 1: Determination of material loss from fabrics during washing ISO/DIS 4484-2: 2023 Textiles and textile products – Microplastics from textile sources – Part 2: Qualitative and quantitative evaluation of microplastics ISO/FDIS 4484-3: 2023 Textiles and textile products – Microplastics from textile sources – Part 3: Measurement of collected material mass released from textile end products by domestic washing method	
Area „soil“			CEN/TC 444 Environmental characterization of solid matrices Task group “Microplastics”

Microplastics : also some protocols from US

US ASTM

- ASTM D8332 − 20 : Standard Practice for Collection of Water Samples with High, Medium, or Low Suspended Solids for Identification and Quantification of Microplastic Particles and Fibers
- ASTM D8333 − 20 Standard Practice for Preparation of Water Samples with High, Medium, or Low Suspended Solids for Identification and Quantification of Microplastic Particles and Fibers Using Raman Spectroscopy, IR Spectroscopy, or Pyrolysis-GC/MS

Southern California Coastal Water Research Project Authority

- Standard Operating Procedures for Extraction and Measurement by Infrared Spectroscopy of Microplastic Particles in Drinking Water
- Standard Operating Procedures for Extraction and Measurement by Raman Spectroscopy of Microplastic Particles in Drinking Water
- SCCWRP SOP for all waters, sediments to be issue under several weeks

Microplastics method in clean water : key messages

- Only reliable identification at low numbers and low size but time-consuming through Spectroscopy : Micro-Raman or Micro-FTIR
- As MPs ubiquitous in lab environment (air, consumable,…), difficult to have lab blank/reference non polluted
- Numerous parameters, complex settings, fine tuning and quality criteria for obtaining expressing reliable results
- Standard will impose laboratories to verify and validate their method implementation. For Spectroscopy measurement, it will be done according to size measurement, right identification at claimed size, recovery rate, limit of reporting and automatic settings.

- No regulation yet, but new EU Directive for Drinking Water, as California State Water Resources Control ask for monitoring
- No standardized analytical methods yet but strong contribution to an on-going process for water method standardization at AFNOR (FR) and ISO Level (ISO 16094-2:2023 & ISO 16094-3:2023) under Vienna agreement.
- Some protocols have been also been issued in US by ASTM & SCCWRP

THANKS to Alina Maltseva & Ulrike Braun