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Analysis of microplastics as emerging contaminants using single particle ICP-MS

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Goal: To demonstrate the applicability of the Thermo Scientific™ iCAP™ RQ ICP-MS in conjunction with the scQuant plug-in for Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution Software for the characterization of microplastics in environmental water samples.

Keywords: Environmental analysis, mass distribution, microplastics, pollution, single particle, ICP-MS

INTRODUCTION

Plastics are widely used in many products, electronics and packaging (foils, bottles, foams, etc). Due to their physical formation, plastics often have high strength and are resistant to corrosion. As the overall amount of plastics produced globally continues to increase, so does the amount of waste produced. As a result of incorrect disposal of many plastic products, pollution has become a major threat to the environment.

A surprisingly large amount of the environmental contamination observed today arises from less immediately obvious anthropogenic sources, such as rubber particles from tire wear and other microscale plastic debris, which is then subsequently mobilized. A study conducted in 2019 estimated the contribution of microplastic particles washed off by rain into storm drains in the San Francisco Bay Area to exceed 300 times the amount of plastic pollution that enters the environment through sewers and sewage plants.[1]

Although there is no guidance for a maximum contaminant level yet, the United States Environmental Protection Agency (EPA) has released its Microplastic Beach Protocol, containing a detailed work instruction for volunteers to scientifically collect and analyze microplastic pollution (here with typical sizes in the millimeter range) in the environment.[2] This direction has accelerated focus on evaluating the extent of pollution of the environment with microplastics, as well as on research into the degradation of these particles over time and possible ways for protecting environmental and human health on a larger scale.

Other analytical techniques, such as RAMAN spectroscopy and pyrolysis gas chromatography-mass spectrometry (py-GC-MS) have been described for the analysis of microplastics,[3] allowing the identification of the polymeric backbone of the material and the nature of the compound. However, there are a couple of challenges that remain, such as the need for complex sample preparation as the microplastics must be collected from the original sample and concentrated prior to analysis. This means that results can take a long time to generate and often large sample volumes are required.

Inductively coupled plasma mass spectrometry (ICP-MS) has been used for the analysis of nanoparticles, allowing the investigation of a reasonably high number of particles in a short time, and providing an overview on the average size and number composition of a sample. Since there are almost no trace metals intrinsically present in microplastics, direct ICP-MS analysis was for long not considered feasible, as analysis of carbon by ICP-MS is considered difficult due to the low ionization yield for this element and elevated backgrounds due to the ubiquitous presence of carbon as CO_2 in the surrounding air and dissolved in all aqueous solutions prepared for analysis. However, for particles of larger size, such as microplastics, the use of ICP-MS operated in time-resolved mode for carbon measurement presents a promising alternative for studying the rate of plastic degradation in environment pollution.[4]

This application note demonstrates how the iCAP RQ ICP-MS was used to detect the carbon in microplastics in both standard solutions as well as in a drinking water sample fortified with microplastic material.

EXPERIMENTAL

Experimental optimization of instrument parameters

An iCAP RQ ICP-MS was used for all measurements. The sample introduction system consisted of a dedicated nebulizer and spray chamber combination (CytoNeb and CytoSpray, Elemental Scientific, Inc., Omaha, NE, USA). The modified sample introduction system also featured a one-piece quartz torch with a 2.0 mm i.d. quartz injector. The overall purpose of this modification is to improve the transport efficiency for larger particles, which would otherwise not be able to reach the plasma. The sample solution was delivered using a syringe pump (Fusion 100-X, Chemyx, Inc., Stafford, TX, USA) and a 500 µL glass syringe (Hamilton Company, Reno, NV, USA) as low sample flow rates (in this case 20 µL·min^{-1}) are required to ensure optimal performance of the sample introduction system. The syringe pump was controlled using a dedicated Qtegra ISDS Software plug-in. To further improve sensitivity, the high sensitivity skimmer cone insert was selected for this application. To reduce potential background signals, an argon gas filter (Entegris, Inc., Billerica, MA, USA) was installed on the argon gas supply of the instrument to remove any carbon containing impurities. Pure oxygen was investigated for use as a reactive gas in the QCell collision/reaction cell (CRC). Table 1 gives an overview of the full configuration of the system.

Table 1. Instrument configuration and operating parameters

Parameter	Value
Spray chamber	ESI CytoSpray spray chamber
Torch	ESI one-piece quartz torch with 2.0 mm i.d., injector
Nebulizer	ESI CytoNeb
Syringe pump speed	Flow rate 20 µL·min^{-1}
Interface	Platinum sampler and platinum tipped skimmer cone with high sensitivity skimmer cone insert
Plasma power	1,550 W
Nebulizer gas	0.4 L·min^{-1}
Additional argon gas	0.5 L·min^{-1}
Sample depth	7 mm
QCell setting	O_2 CCTS
Cell gas flow	100% O_2, 0.3 mL·min^{-1}
CR bias	-6.3 V
Q3 bias	-12 V
Scan settings	0.01 s dwell time
Quadrupole	High resolution (0.3 amu)
Isotope monitored	$^{12}C^+$

Table 1. Instrument configuration and operating parameters (continued)

Parameter	Value
Sample uptake	90 s
Wash time	10 s
Sample analysis duration*	30–120 s

*Sample time was varied to ensure good counting statistics with sample including particles.

Data acquisition and data processing

The Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution™ (ISDS) Software with the scQuant data evaluation plug-in was used to create LabBooks for sample analysis, data acquisition, processing, and reporting. With the scQuant plug-in, the particle transport efficiency, mass and size distribution were automatically calculated.

Sample preparation

Precleaned polypropylene bottles were used for the preparation of all blanks, standards, and samples. The bottles were rinsed with ultrapure water (18.2 MΩ·cm) and left to dry in a laminar flow clean hood before use.

All blanks and calibration standards used for the determination of the detection sensitivity were prepared from ultrapure water using IPA (Sigma-Aldrich, semiconductor grade) as a carbon source. The transport efficiency (i.e., fraction of particles that are able to pass through the sample introduction system and finally be measured in the plasma) is a key parameter for the correct estimation of the elemental mass per particle and the particle number concentration in the sample. To assess the transport efficiency of the combined system, a 60 nm gold nanoparticle standard, RM 8013 (National Institute of Standard and Technology (NIST)), was used. Although significantly smaller compared to the microplastic particles under investigation here, the aforementioned nanoparticles are well studied and frequently applied to determine the transport efficiency for discrete entities using ICP-MS. Whereas a standard sample introduction system commonly used in ICP-MS, consisting of a quartz nebulizer and a cyclonic spray chamber, would typically only transmit around 5% of these particles to the plasma for ionization, the sample introduction system used in this study showed a higher transport efficiency of up to 48%. This improvement was due to the high efficiency of the nebulizer, in combination with the reduced sample flow rate of only 20 $\mu L \cdot min^{-1}$.

Further transport efficiency standard solutions containing microplastic particles were prepared using commercially available polystyrene beads with 2 and 5 µm nominal diameter (Sigma-Aldrich) to assess transport of actual microplastic particles in the system. These solutions were further diluted using ultrapure water.

In addition, potable and surface water samples were collected in Bremen, Germany for analysis as real samples. The surface water sample originated from a pond and contained partly undissolved matter. For purification prior to analysis, 5 mL HNO_3 and 2 mL H_2O_2 were added into 50 mL of pond water and left to react for one week. After this period, the sample was centrifuged and diluted 20x with ultrapure water. All samples were sonicated for 1 minute immediately before preparation of a diluted sample and again prior to analysis to resuspend potentially settled particles.

RESULT AND DISCUSSION

Detection of carbon in microplastics

To monitor microplastics using ICP-MS, there are two options. Since ICP-MS is an analytical technique developed to monitor trace elements, and in particular metals, with high sensitivity, metal impurities accumulated in the microplastics could be a way to visualize the particles. However, impurities must be present at a certain level, and contaminants would need to be known as associated exclusively with microplastic particles prior to analysis to allow for targeted screening. Another option is to monitor carbon as the main constituent directly, and hence expand the access of the technology not only to potentially weathered samples but also to "uncontaminated" particles. In general, the analysis of carbon using ICP-MS is one of the most challenging applications because of the high ionization energy (11.26 eV), and contamination from dissolved carbon in the measured solutions (as carbonate or carbon dioxide) and gaseous carbon compounds in the ambient atmosphere surrounding the plasma and the argon gas of the ICP-MS plasma source. In addition, real samples, derived in an environmentally focused study may induce specific interferences, such as $^{11}B^{1}H^{+}$ or $^{24}Mg^{++}$. Regardless of these difficulties, the direct determination of microplastics using the carbon signal has been published recently.[5]

As is also the case for the analysis of nanoparticles, it is important to maximize signal to noise (S/N) and minimize backgrounds to confidently distinguish particle signals. Although collision/reaction cell technology in ICP-MS provides many potential solutions to minimize background interferences, the use of pure O_2 gas was selected in this work.

Figure 1 shows a comparison of the (blank subtracted) signal-to-noise ratio obtained with different O_2 gas flow rates and various isotopes and product ions. The best sensitivity was obtained using a flow rate of 0.3 mL·min^{-1} O_2 flow rate in combination with the detection of $^{12}C^{+}$ (the most abundant nuclide, with 98.9% natural abundance).

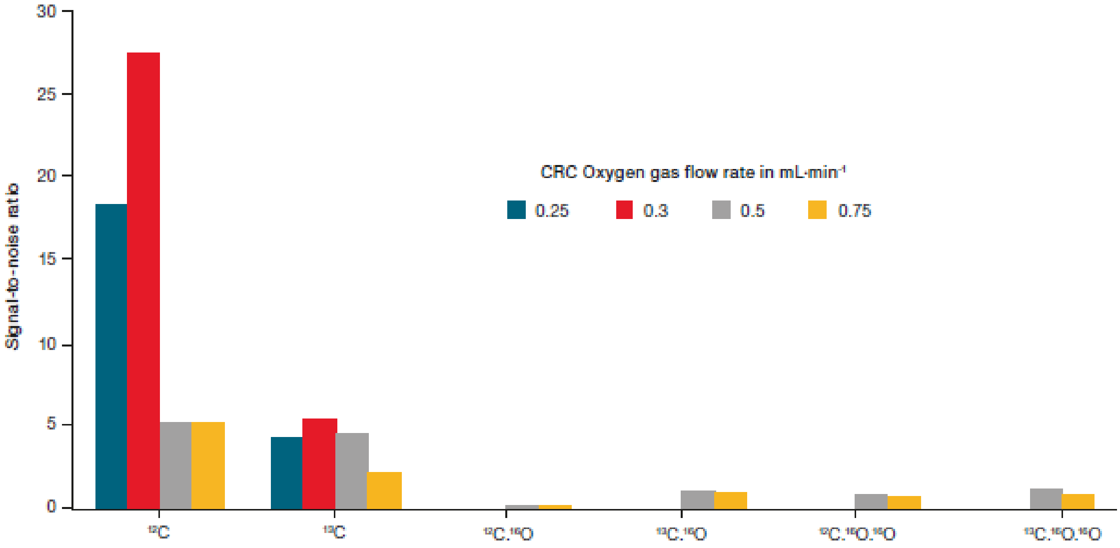
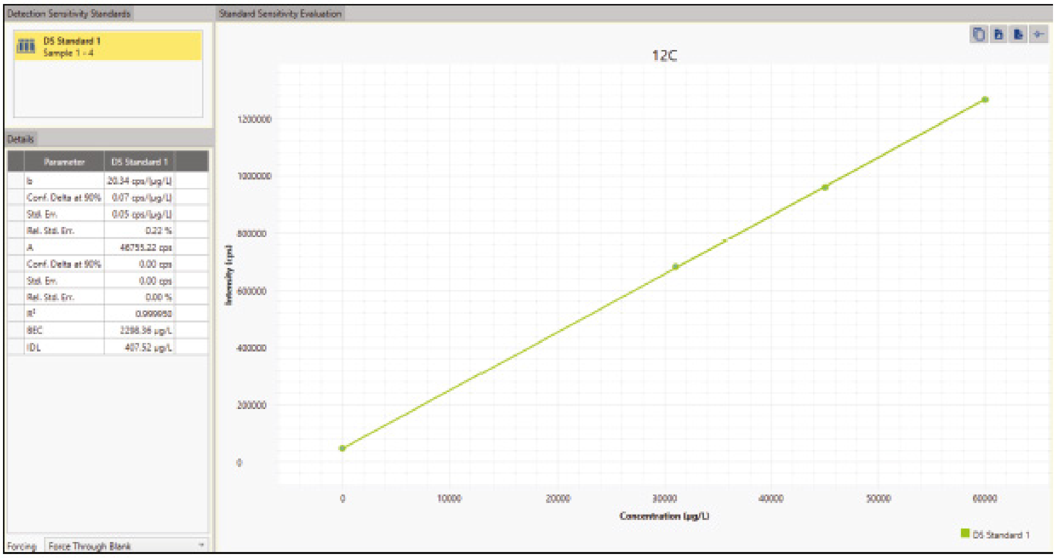


Figure 1. Comparison of carbon S/N for five carbon isotope/molecular with different CRC O_2 gas flow rate.

Using the optimized settings, a calibration curve for carbon was generated by running the respective standard solutions over a range of 31 to 60 mg·L^{-1}. The calibration curve produced (as shown in Figure 2) showed excellent linearity with a coefficient of determination (R^2) value of better than 0.99995, a background equivalent concentration (BEC) of 2 mg·L^{-1} for carbon, and an instrumental detection limit (IDL) of 0.4 mg·L^{-1}.



Parameter	DS Standard 1
b	20.34 cps/(µg/L)
Conf. Delta at 90%	0.07 cps/(µg/L)
Std. Err.	0.05 cps/(µg/L)
Rel. Std. Err.	0.22 %
A	46755.22 cps
Conf. Delta at 90%	0.00 cps
Std. Err.	0.00 cps
Rel. Std. Err.	0.00 %
R²	0.999950
BEC	2298.36 µg/L
IDL	407.52 µg/L

Forcing: Force Through Blank

Figure 2. Calibration curve for $^{12}C^+$ in CCTS O_2 mode on the iCAP RQ ICP-MS.

Analysis of polystyrene standards as a surrogate for microplastics

To investigate whether the detection of microplastic particles is feasible using the proposed approach, 2 and 5 µm polystyrene beads were diluted and analyzed. As can be seen from the data shown in Figure 3, a series of signals were observed, each of them corresponding to a single microplastic particle ionized in the plasma.

In both cases, the overall number of signals increases with decreasing dilution as more particles can enter the plasma and become ionized. The quantitative assessment of the particle number concentration correlates well with the dilution factor, as can be seen in Table 2.

Figure 3 shows that the different particle sizes show different average signal intensities, which is expected as each 5 µm particle contains a significantly higher amount of carbon than the 2 µm particles. As both particle size stock solutions contain the same microplastic mass concentration of 10% (w/w), this translates to a higher number concentration for the 2 µm particles and therefore different dilution factors had to be applied to achieve a comparable particle number concentration in the measured solutions. From the data presented above, the transport efficiency for the different polystyrene beads was estimated to be around 4.19% for 5 µm sized particles and 30.08% for 2 µm sized particles. There is a size effect on the transport efficiency by counting, most likely due to static effects in the sample transport tubing, so care must be taken to apply the correct transport efficiency by counting to the different fractions so that an accurate number concentration is estimated.

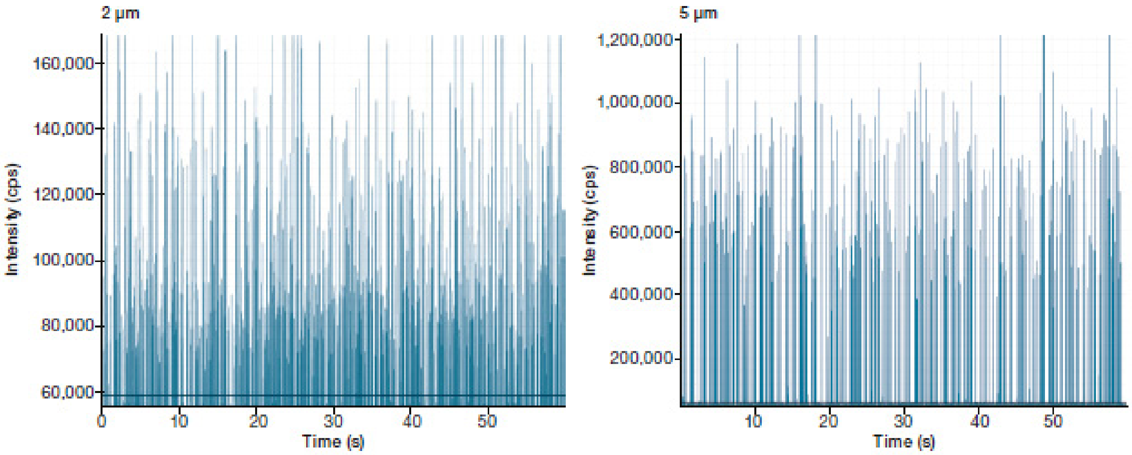


Figure 3. Raw signal intensity data for 2 µm (200,000-fold dilution) and 5 µm (5,000-fold dilution) polystyrene beads.

Table 2. Particle number concentration results with different dilution factors

Sample	Dilution factor	Particle number expected [n·mL^{-1}]	Particle number concentration [n·mL^{-1}]	Particle transport efficiency [%]
Polystyrene beads, 5 µm	5,000	291,026	18,968	4.19
	10,000	145,513	10,108	
Polystyrene beads, 2 µm	100,000	227,364	20,717	30.08
	200,000	113,682	10,658	

Figure 4 shows the workflow of how individual particles are measured and evaluated in the dedicated scQuant plug-in. Image A in the figure shows the raw data as it is acquired by the instrument. Every individual polystyrene particle that is introduced to the plasma (and subsequently becomes ionized and transferred into the mass spectrometer) causes a short transient signal. Using the applied setting for the dwell time in combination with an appropriate dilution of the signal allows individual particles to be fully resolved and their mass to be determined, as well as the number of particles per unit time to be counted. The raw data is converted to a signal distribution plot (B in Figure 4), from which the signal range that corresponds to the particle fraction can be isolated from the background. Once this is accomplished, all selected signals are evaluated, and a mass distribution plot is created. In this step, the measured signal intensity is converted into the mass of the particle using the slope of the calibration curve and the sample flow rate to the plasma. Particle mass and number concentration can also be retrieved from this plot.

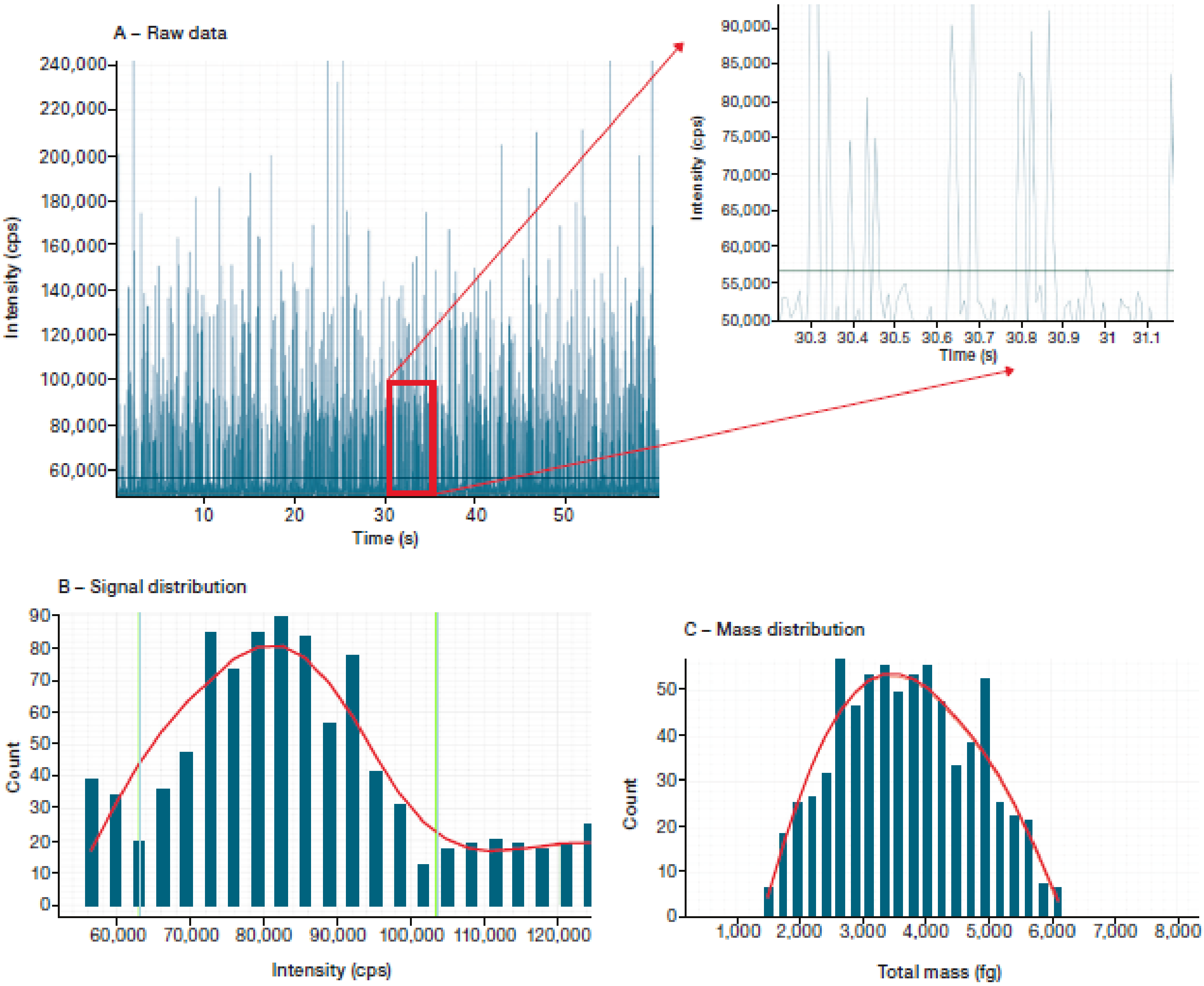


Figure 4. The workflow for the data evaluation with 2 µm polystyrene beads standard solution using scQuant plug-in.

To finally confirm discrete microplastic particles as the origin of the observed signals, the average signal intensity observed for both particle sizes investigated in this study was calculated and compared. Assuming a spherical shape of the particles, the approximate number of atoms per particle should change following the nominal size relation. Note that as the radius contributes in the third power to the volume of a sphere, the absolute signal intensity should change as per the following calculation:

$$\text{Volume } 5\ \mu m / \text{Volume } 2\ \mu m = (3/4\pi(2.5)^3) / (3/4\pi(1.0)^3) = 15.625/1 = 15.625 \qquad \text{Equation 1}$$

Table 3 shows the intensity of ten selected signals from the measurement of polystyrene microplastics monitoring $^{12}C^+$. The average intensity ratio observed between particles with 2 and 5 µm nominal diameters was determined to be 15.42 ± 0.17, which is very close to the theoretical result determined from Equation 1.

Table 3. Intensity ratio result with 2 and 5 μm polystyrene microplastics monitoring $^{12}C^+$. All intensities are background subtracted.

	Diameter of microplastics	
Signal	**2.0 μm**	**5.0 μm**
1	59,441	916,027
2	59,240	923,348
3	59,140	910,275
4	59,843	905,046
5	59,541	923,348
6	59,642	911,844
7	59,541	916,550
8	59,039	929,101
9	59,843	925,963
10	59,742	918,642
Average	59,501	918,014
SD	285	7,569
Experimental ratio	15.42 ± 0.17	
Theoretical ratio	15.62	

Analysis of unknown environmental samples

To illustrate the use of this method for the analysis, two different environmental water samples (potable water and pond surface water) were screened for the presence of microplastics. A challenge in the analysis of real water samples, compared to standard solutions prepared in ultrapure water, is the higher amount of dissolved carbon dioxide (CO_2), causing an increase of the background signal observed in the measurement, and potentially overlaying the microplastic particles present. This is highlighted in the mass distribution plot shown in Figure 5, which shows a large number of signals counted on the left of the distribution that result from the carbon background. To reduce the background on $^{12}C^+$, both water samples were diluted with ultrapure water by a factor of 5 before the analysis. Note that this dilution only leads to a reduction of the number concentration of any particles; the signal intensity derived from individual particles will remain unchanged, so that they can still be detected despite the larger dilution of the sample. To obtain similar counting statistics (to ensure that a representative number of particles are counted in a sample), the overall measurement time can be extended if required. During the analysis of both water samples, however, no detectable signals for microplastics were found, so the polystyrene beads previously investigated were used to perform a spike recovery experiment.

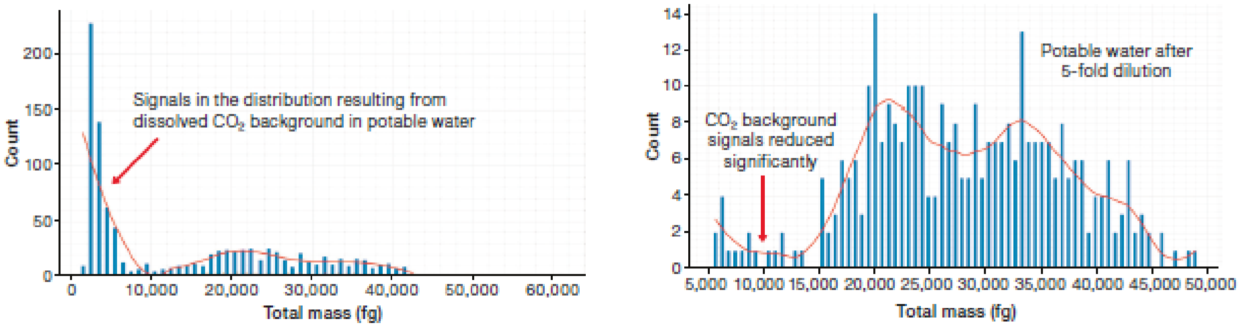


Figure 5. Mass distribution result of potable water with and without dilution, showing reduction of signals resulting from background CO_2.

Table 4 shows the analysis results for the 5 µm polystyrene beads spike recovery test in both natural waters. A 5 µm spike recovery result for five-fold diluted potable water of 84.9% was obtained, while approximately 50% for pond water was achieved. Diluting the sample improves the accuracy of the particle number analysis, which is one of the key steps required to ensure accurate analysis of microplastics. Additionally, although not shown here, longer sample analysis times allow more particles to be collected, thereby improving counting statistics and data quality.

Table 4. Spike recovery results for two environmental water samples

	5-fold dilution		**20-fold dilution**		**5 µm spike in ultra pure water (reference value)**
	Potable water	**Spiked potable water**	**Pond water**	**Spiked pond water**	
Durations (s)	120	120	60	60	30
Mean (fg)	60,225 ± 7,538	57,822 ± 8,920	60,865 ± 12,280	59,576 ± 10,676	62,146 ± 10,995
Number (particle)	3	153	5	46	48
Number per volume (particle·mL^{-1})	149	13726	415	7652	15973
Spiked particle recovery (%)	84.9%		45.3%		-

CONCLUSIONS

A highly sensitive method for microplastics analysis using the iCAP RQ ICP-MS equipped with scQuant software was developed and applied to the analysis of two environmental water samples. The analytical method was rigorously tested for performance, and the results obtained clearly demonstrated the following analytical advantages:

- Interference-free analysis was achieved using high-performance collision / reaction cell operation with high mass resolution (0.3 amu). By minimizing the carbon background during sampling, a carbon BEC of 2 mg·L^{-1} can achieved, allowing the use of $^{12}C^+$ for the direct ICP-MS analysis of microplastics in environmental samples.
- Polystyrene beads could be detected via single particle events and, from the data, the mass distribution for 2 and 5 µm particle sizes could be determined.
- The scQuant evaluation tool offers automatic calculation of the particle transport efficiency, mass and signal distribution enabling effective analysis of microplastic particles in the samples studied.

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