Single Particle ICP-TOFMS with Online Microdroplet Calibration: A Versatile Approach for Accurate Quantification of Nanoparticles, Sub-Micron Particles, and Microplastics in Seawater

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Single particle inductively coupled plasma time-of-flight mass spectrometry (spICP-TOFMS) is a powerful analytical technique for quantifying elements in nanoparticles and microparticles; however, like most ICP-MS based measurements, matrix effects can be a major challenge for accurate quantification in spICP-MS. Here, we report the use of online microdroplet calibration to overcome extreme matrix effects observed for the analysis of nanoparticles and microparticles in seawater. In online microdroplet calibration, particle-containing samples are introduced into the ICP along with monodisperse microdroplets containing known element mass amounts. The microdroplet standards, which experience the same plasma conditions as analyte particles, are used to measure matrix-matched absolute element sensitivities. With online microdroplet calibration, one multi-elemental standard can be used to determine the element mass amounts in diverse types of analyte particles independent of the sample matrix.

We evaluate the matrix tolerance of spICP-TOFMS with online microdroplet calibration through the analysis of metal nanoparticles, polystyrene microplastic beads doped with rare-earth elements, and metal-oxide sub-micron particles in artificial seawater. Our results demonstrate mass recoveries of 98−90% for the analysis of individual gold NPs in ultrapure water to 99% seawater. In the analysis of food grade TiO_2 submicron particles, accurate Ti-mass per particle is determined with matrix-caused signal attenuation up to 80% in pure seawater matrix. We also demonstrate accurate diameter determinations of individual 3.4 µm polystyrene beads at concentrations of up to 80% simulated seawater. Furthermore, simultaneous and accurate quantification of rare-earth elements in the polystyrene beads is achieved.

Introduction

The widespread contamination of aquatic ecosystems by microplastics, nanoparticles, and other pollutants has become a significant environmental concern, and the need for new and innovative methods to measure and quantify these pollutants is becoming increasingly urgent.[1-2] Nano- and microplastics are major contributors to pollution in ocean environments. One model estimates that there are 0.27 tons of plastic litter in the ocean, with microplastics accounting for 92% of the total plastic particles or 13% of the weight.[3] Heavy metals such as Pb, Cd, As, and Hg have been shown to adsorb to microplastics, which are potential vectors for these metals in the marine environment.[4-6] Additionally, numerous studies have investigated the effects of nano pollution on aquatic organisms and have shown that metallic nanomaterials can disrupt or alter biological processes in model organisms.[7-9] As such, the development of accurate and sensitive analytical methods for measuring these pollutants is crucial.

Unfortunately, the quantification of nanoparticles (NPs) and microparticles (µPs) in environmental matrices like seawater is challenging with current methods.[1] Additionally, the dilute nature of anthropogenic NPs and pollutant microplastics makes some low throughput methods, such as microscopy, less useful for studying particle populations.[10] Single-particle inductively-coupled plasma mass-spectrometry (spICP-MS) has been developed as a high throughput method for the quantification of element mass amounts in individual particles and particle number concentrations (PNCs). With spICP-MS, a dilute particle suspension is introduced into an ICP such that only one particle enters the plasma at a time. These discrete particles are then vaporized, atomized, and ionized in the ICP, and produce clouds of ions that—after being extracted into the mass analyzer—have temporal durations of ~200 to 500 µs. The mass analyzer is operated in high-time resolution mode and the discrete ion clouds are separated according to ions' mass-to-charge (*m/z*) ratios such that element- and isotope-specific transient signals are recorded for each particle. spICP-MS has been used to determine the mass and number concentrations of particles ranging in size from 5 nm – 1 µm; however, information regarding elemental composition at the single particle level is limited by the number of elements that can be simultaneously measured.[11-16]

In this research, we use a time-of-flight mass spectrometer for spICP-MS analysis. With spICP-TOFMS, ions across the entire atomic mass range—from ^{6}Li to ^{238}U—can be analyzed quasi-simultaneously with sufficient time resolution to detect particle-produced transient events.[17-19] Number concentrations analyzed by spICP-MS are in the range of 10^3-10^6 particles mL^{-1}, which corresponds to the detection of tens to thousands of particles per minute and makes the approach well suited for the measurement of dilute samples. spICP-MS has been extensively applied to the measurement of metal-containing nanoparticles in freshwater bodies of water such as rivers[20-23] and lakes.[21, 24] However, seawater remains a challenging matrix for ICP-MS measurements due to the high salt content, which can change the ionization efficiency of atoms in the plasma.[25-29] To date, most spICP-MS measurements of particles in seawater have involved the removal or dilution of the sample matrix prior to measurement.[30-31] While sample dilution was effective for some spectral interferences, it still required the preparation of matrix-matched standards due to the signal attenuation.

Commonly, element mass amounts are calibrated in spICP-MS measurements with an approach called the particle size method,[32] which involves separate measurements of various dissolved-element standards and a reference particle standard. While the particle size method is effective for the calibration of particle mass and size in simple samples, the application of the method for the analysis of particles in difficult matrices is a challenge.[33] Accuracy of the particle size method depends on externally determined absolute sensitivity values that, in turn, depend on external transport efficiencies (η_{neb}) and plasma uptake (q_{plasma}) values. Thus, with the particle-size calibration method, any change in nebulized aerosol transport efficiency or absolute sensitivities–either from a changing sample matrix or from drift–will result in an inaccurate mass quantification. To improve the accuracy of particle quantification, approaches based on as internal standardization,[34-35] standard addition,[36] isotope dilution,[37-38] and matrix-matched standards[39] have been used. Here, we demonstrate the used of online microdroplet calibration[40-42] (Fig. 1) as an accurate calibration strategy for spICP-TOFMS that eliminates the need for exhaustive sample preparation and matrix removal procedures.

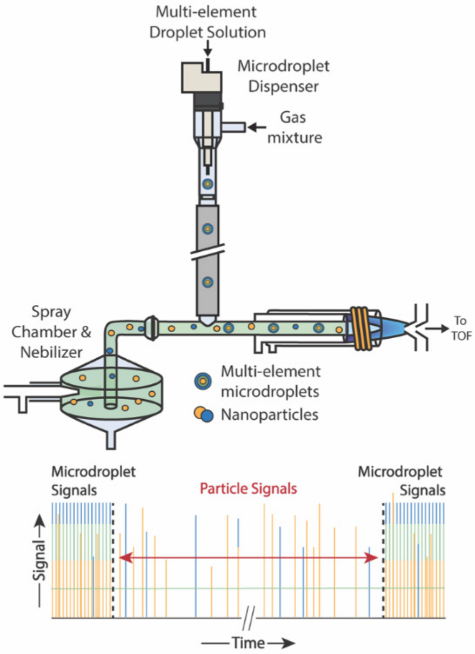

Figure 1. Schematic diagram of online microdroplet calibration system and data structure.

In online microdroplet calibration, monodisperse microdroplets (~70 µm, ~180 pL) containing a known concentration of a calibration solution are generated and introduced into the ICP with a dual-sample introduction system. Microdroplet standards are introduced along with particle-containing samples and are used to determine the absolute sensitivities for each element present in the droplets for each measurement. Technical details of the online microdroplet approach can be found in previous publications.[41-42] Because microdroplet standards are introduced into the plasma along with nebulized sample aerosols, the droplets experience the same plasma conditions as the analyte particles, which enables automatic matrix-matched calibration. If a sample matrix causes signal attenuation or enhancement from analyte particles, then the droplet-produced signals will be similarly attenuated or enhanced and calibration will be accurate. Online microdroplet calibration does not require the creation of matrix-matched particle and dissolved-element standards, which significantly improves the throughput of the measurements compared to conventional spICP-MS calibration strategies, e.g. the particle-size method. Moreover, since calibration is done for each measurement individually, online microdroplet calibration accounts for changes in instrument performance with time, i.e. drift, and corrects for sample-to-sample matrix variations. This makes online microdroplet calibration ideal for sample sets that may have varied or unknown matrix compositions. In this work, we present the use of online microdroplet calibration for the measurement of metal (Ag and Au) NPs, TiO_2 sub-micron particles, and microplastics in artificial seawater.

Experimental

Instrumentation

An icpTOF-S2 instrument (TOFWERK AG, Thun, Switzerland) equipped with a microFAST MC syringe-based autosampler (Elemental Scientific, Omaha, NE, USA) and the online-microdroplet calibration setup[42] was used for all measurements. The instrument was tuned daily. Highly abundant matrix ions were notched using the instrument's notch filter to prevent detector saturation; additionally, the collision cell was used with H_2 and He gas. Detailed instrument parameters are provided in Table S1. Samples were introduced using a PFA MicroFlow pneumatic nebulizer (Element Scientific Inc., Omaha, USA) into a cyclonic spray chamber (Glass Expansion, Hawthorn, Victoria, Australia) at a flow rate of 45 µL min^{-1}. Microdroplets were generated using a microdroplet dispenser head (50 µm nozzle diameter, AD-KH-501-L6 with control unit MD-E-3011-131, Microdrop Technologies, Norderstedt, Germany). They were introduced into a 30 cm length of aluminum tubing at 40 Hz. A mixture of ~400 mL min^{-1} helium and ~10 mL min^{-1} argon gas was introduced to the top of the tube to transport and desolvate the microdroplets. The dried droplet residues were mixed in a t-piece with the analyte-particle-containing aerosol from the spray chamber and then both microdroplet standards and analyte particles were introduced in a combined gas stream into the central channel of the ICP torch.

Materials

PVP (polyvinylpyrrolidone)-coated gold nanoparticles (Au NP, diameter = 102 ± 8 nm) and silver nanoparticles (Ag NP, diameter = 81 ± 6 nm) were obtained from nanoComposix, now Fortis Life Sciences (San Diego, CA, USA), and E171 TiO_2 particles (d50, 800 nm) were purchased from Sigma-Aldrich, Burlington, MA, United States. The Ag NPs were measured with SEM and determined to have an actual diameter of 72 ± 8 nm (see SI). Multi-element doped polystyrene (PS) beads (EQ Four Element Calibration Beads) were purchased from Standard BioTools Inc., formerly Fluidigm (San Francisco, California, USA). Simulated sea salt was purchased from Lake Products Company LLC (Florissant, MO, USA) and prepared to standard ASTM D1141-98(13) for the preparation of substitute ocean water (Table S4). Multi-element calibration solutions for online microdroplet calibration were prepared with concentrations ranging from 50 to 300 ng mL^{-1} from single-element standards (High-Purity Standards, South Carolina, USA), except for the carbon standard, which was prepared from tartaric acid (Fisher Scientific, Waltham, MA, USA) to a concentration of 237 µg C mL^{-1}. Standards were diluted gravimetrically using 1% (v/v) sub-boiled HNO_3 in ultrapure water or a mixture of 1% (v/v) HNO_3 and 3% (v/v) HCl (TraceSelect Grade, Honeywell-Fluka, Charlotte, NC, USA) for solutions containing gold and silver.

Data processing

All ICP-TOFMS data was collected using TofDAQ Recorder and mass spectral peak integrations were done with TofDAQ viewer (Tofwerk AG, Thun, Switzerland). Here, we refer to all icpTOF signals as TofCounts (TofCts) because signals provided by the icpTOF instrument are estimated counts from digitized analog signals from the detector, rather than true counts.[43] Time traces of intensity (i.e. TofCts) for all *m/z* values of interest were processed by our in-house "TOF Single-Particle Investigator" (TOF-SPI) program, written in LabVIEW (LabVIEW 2018, National Instruments Corp., TX, USA) for spICP-MS data analysis. This program is used to calculate background signals and single-particle critical values ($L_{C,sp}$) for all elements of interest, determine absolute sensitivities, background subtract data, correct split-particle events, and quantify element masses in single particles. Critical values were calculated according to compound Poisson distributed ICP-TOFMS data, as previously described.[43-44] Mass amounts of elements in each particle were determined with TOF-SPI, and then reported diameters were calculated by assuming the particles were spherical in shape and had the same density as the bulk material (see Table S4). Figures were made in OriginPro 2019b (OriginLab Corporation, Northampton, MA, USA), and the final figures were assembled in Adobe Illustrator 2021 (Adobe, San Jose, CA, USA).

Results and Discussion

To evaluate the performance of online microdroplet calibration for the quantification of nano- and microparticles in a seawater matrix, Ag and Ag NPs, TiO_2 sub-micron particles, and microplastic REE-doped polystyrene beads were spiked into varying concentrations of simulated seawater. Online microdroplet calibration is advantageous for this type of analysis since it is automatically matrix-matched, so it provides an accurate and high-throughput method for measuring NPs, sub-µm particles, and microplastics. All measurements done with microdroplet calibration are compared to the same measurement done without a matrix matched calibration. Because high salt-content matrices can lead to sedimentation (i.e. salting out) of suspended nanoparticles, we did not quantify particle number concentrations in these samples. In our measurements, we did not observe evidence of agglomerated particles in suspension, such as dimer-particle events.

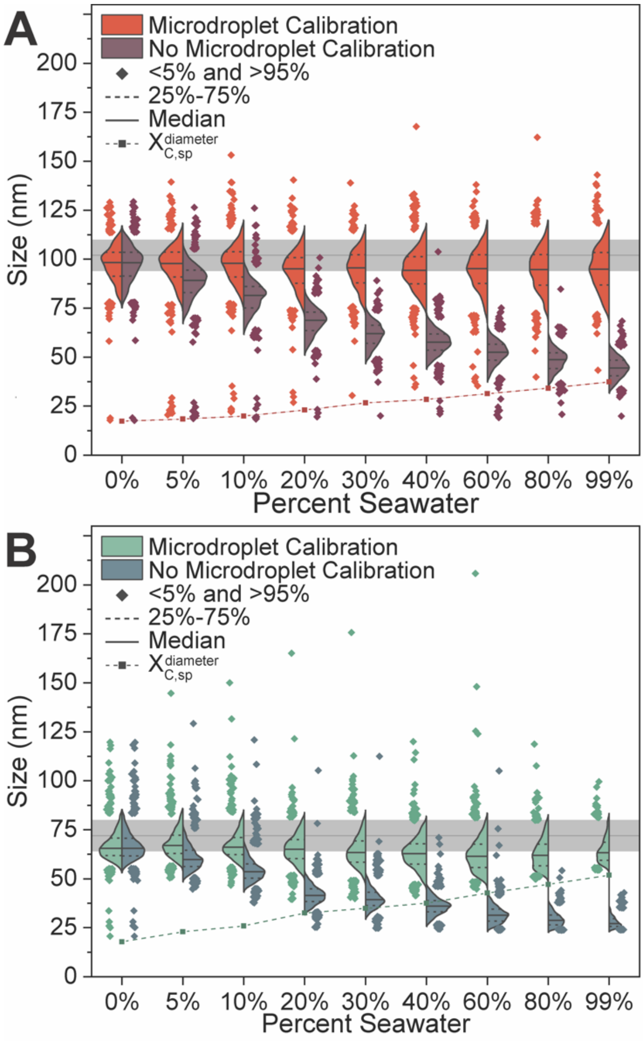

Figure 2. Violin plots showing the distributions of 102 ± 8 nm Au NPs (A) and 72 ± 8 nm Ag NPs (B) in varying concentrations of simulated seawater with and without online microdroplet calibration. The solid grey bar is the nanoparticle size as determined with SEM.

Gold and Silver Nanoparticles in Seawater

Single metal gold and silver nanoparticle standards (102 ± 8 nm and 72 ± 8 nm, respectively) were spiked into varying concentrations (0%-99% v/v) of simulated seawater (Fig. 2). With online microdroplet calibration, the median gold NP diameter was accurately determined in concentrations up to 99% (v/v) seawater, which contains 1.2 wt% Na^+ among other electrolyte ions (see Table S4). In 99% seawater, signals from the nominally 100 nm Au NPs were attenuated 91% compared to their sensitivity in pure water. Likewise, as seen in Fig. S3, the signals from the microdroplets, which contain 9.75 fg Au, were attenuated by 90%. The agreement between the amount of signal attenuation from the droplets and that experienced by the analyte NPs enables accurate determination of element mass in the analyte particles independent of matrix composition.

As seen in Fig 2B, the Ag NPs are sized accurately (the median diameter was within the diameter range measured by SEM) in concentrations up to 30% seawater; however, in greater seawater concentrations, the Ag NPs were undersized. Under sizing of the Ag NPs is partly explained by the fact that the Ag NPs are not perfectly spherical, as assumed when calculating the diameter from the measured mass (see Fig. S2). It has also been shown that Ag NPs can aggregate or dissolve in artificial and filtered seawater samples, though in natural saltwater environments they can be stabilized by extracellular polymeric substances.[30, 45] In addition, as the concentration of seawater increases, the absolute sensitivity decreases due to ionization suppression, and this leads to fewer particles being detected in a given spICP-TOFMS measurement. From pure water to 99% seawater, the sensitivity for Ag goes from 200 to 16 TofCts fg^{-1}, and the critical mass ($\mathrm{X}_{\mathrm{C,sp,Ag}}^{\mathrm{mass}}$) goes from 31 to 764 ag, which corresponds to critical diameters ($\mathrm{X}_{\mathrm{C,sp,Ag}}^{\mathrm{diameter}}$) of 18 to 52 nm. The critical diameter is the minimum detectable diameter by spICP-TOFMS; when this diameter becomes near the diameter range of analyte particles, then fewer particles are measured, which can bias the determined median diameter. At 80% and 99% seawater, the measured Ag NP median diameters increase slightly compared to the determined diameter at 60% seawater; this increase is likely due to elevated $\mathrm{X}_{\mathrm{C,sp,Ag}}^{\mathrm{diameter}}$ at higher salt content. With spICP-TOFMS and online microdroplet calibration, the 72 ± 8 nm Ag NPs were determined to have median diameters of 66 nm in water and 62 nm in 99% seawater; in 99% seawater, 82% Ag mass recovery in individual particles is obtained. Without online microdroplet calibration, the median determined diameter of the Ag NPs is 27 nm in 99% seawater, which is a mass recovery of just 7%.

Altogether, our results show that, with the online microdroplet calibration, measured mass recoveries of Au NPs and Ag NPs are within 90% and 82% for all seawater concentrations. Online microdroplet calibration can be used to compensate for signal suppression up to two orders of magnitude so long as the critical diameter for a measurement doesn’t exceed the diameter of analyte particles.

Microplastics in Seawater

Single-particle ICP-MS has previously been applied to the measurement of microplastic particles;[46-49] however, the approach has not yet been applied for the analysis of microplastics in seawater. Here, we used uniform polystyrene beads doped with rare-earth elements (Ce, Eu, Ho, and Lu) as a model microplastic. The instrument was tuned for low *m/z* sensitivity to allow for the detection of $^{12}C^+$.[50] The polystyrene beads were measured in concentrations of up to 80% (v/v) seawater. The $^{12}C^+$ signal from the beads was detected, and the mass was quantified both with and without microdroplet calibration. Size determinations were calculated using a density of 1.05 g cm^{-1} and a particle stoichiometry of C_8H_8. A particular challenge of carbon particle detection in seawater is that, in addition to reduced sensitivity due to ionization suppression from EIEs, seawater contains carbonate/bicarbonate anions which causes the dissolved carbon concentration to increase with seawater concentration.

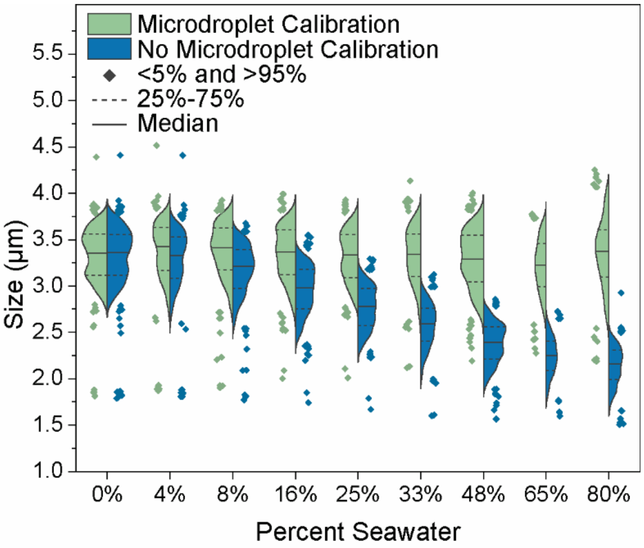

Figure 3. Violin plots of polystyrene bead diameters determined both with (green) and without (blue) online microdroplet calibration.

In Figure 3, we present the determined diameter distributions of the polystyrene beads both with and without online microdroplet calibration as a function of percent seawater. In both ultrapure water and the 80% seawater matrix, the average diameter of the polystyrene beads determined with online microdroplet calibration was 3.4 µm. Without microdroplet calibration, the determined average diameters of the beads decreased to 2.2 µm with an 80% seawater matrix, which is 30% average mass recovery of carbon per particle. The sensitivity for ^{12}C decreases from 27 TofCts pg^{-1} in water to 7 TofCts pg^{-1} in 80% seawater. Microdroplet calibration was able to correct for changes in sensitivity in all the concentrations of seawater tested; however, a higher concentration of seawater does increase the minimum detectable particle size. With a 33% seawater matrix, the ^{12}C sensitivity was 12.6 TofCts pg^{-1} with a critical diameter ($X_{C,sp}^{diameter}$) of 2 µm, i.e. 4 pg of carbon per particle (Fig. S7). Therefore, to maintain the capability to detect microplastics with effective diameters of ~2 µm and above, we suggest that 3-fold dilution of seawater represents a good compromise for the analysis of real samples. This dilution factor prevents substantial sensitivity loss and also avoids excessive dilution of already dilute particle suspensions expected in natural samples.

In addition to measuring ^{12}C signals from the microplastics, with ICP-TOFMS, we can measure all elements in the beads simultaneously with the carbon signals. Since the instrument tuning was optimized for the measurement of low-*m/z* ions, the sensitivities for the rare-earth elements (REEs) decreased more rapidly than that of carbon as the amount of seawater in the matrix increased. As seen in Figure 4, with microdroplet calibration, the measured mass distribution of the REEs in the polystyrene beads was consistently quantified with online microdroplet calibration up to a concentration of ~33% seawater. At a 3-fold dilution of seawater, REE sensitivities as recorded by microdroplets were decreased 75-89% compared to sensitivities recorded in ultrapure water (Fig. S5). Thus, without online microdroplet calibration, the mass recoveries of these REEs would be 10-26%. The mass recoveries of Ce, Eu, Ho, and Lu are 102, 109, 105, and 102% with online microdroplet calibration in the 33% seawater matrix.

At seawater concentrations above 33%, the sensitivity of the REEs is attenuated so much that the critical mass ($\mathrm{X}_{\mathrm{C,sp}}^{\mathrm{mass}}$) for the REEs approaches the true mass distributions of the REEs in the beads. Once $\mathrm{X}_{\mathrm{C,sp}}^{\mathrm{mass}}$ is larger than a significant portion of the true element mass distribution, the recorded mass distribution is not representative of the original sample. Particles with lower masses of the REEs are not measured, and so the recorded mass distribution is truncated at the low mass side and thus biased towards high mass particles. In Fig. 4, we indicate the point at which $\mathrm{X}_{\mathrm{C,sp}}^{\mathrm{mass}}$ exceeds the first quartile of the mass distribution recorded in ultrapure water for each of the elements. At higher concentrations of seawater (48-80% v/v), the recorded mass distributions of all the REEs trend upward. With increasing seawater content in the samples, fewer and fewer particles are measured, and this causes outlier measurements, such as double events, to have a greater impact on the overall distribution. The biased recorded mass distributions of REEs at high seawater concentrations is not a limitation of online microdroplet calibration; the masses of elements in the particles are still accurately quantified. Rather, biased recorded mass distributions are inherent to spICP-MS measurements; only improving absolute sensitivity, and thus lowering $\mathrm{X}_{\mathrm{C,sp}}^{\mathrm{mass}}$ values, will overcome this bias.

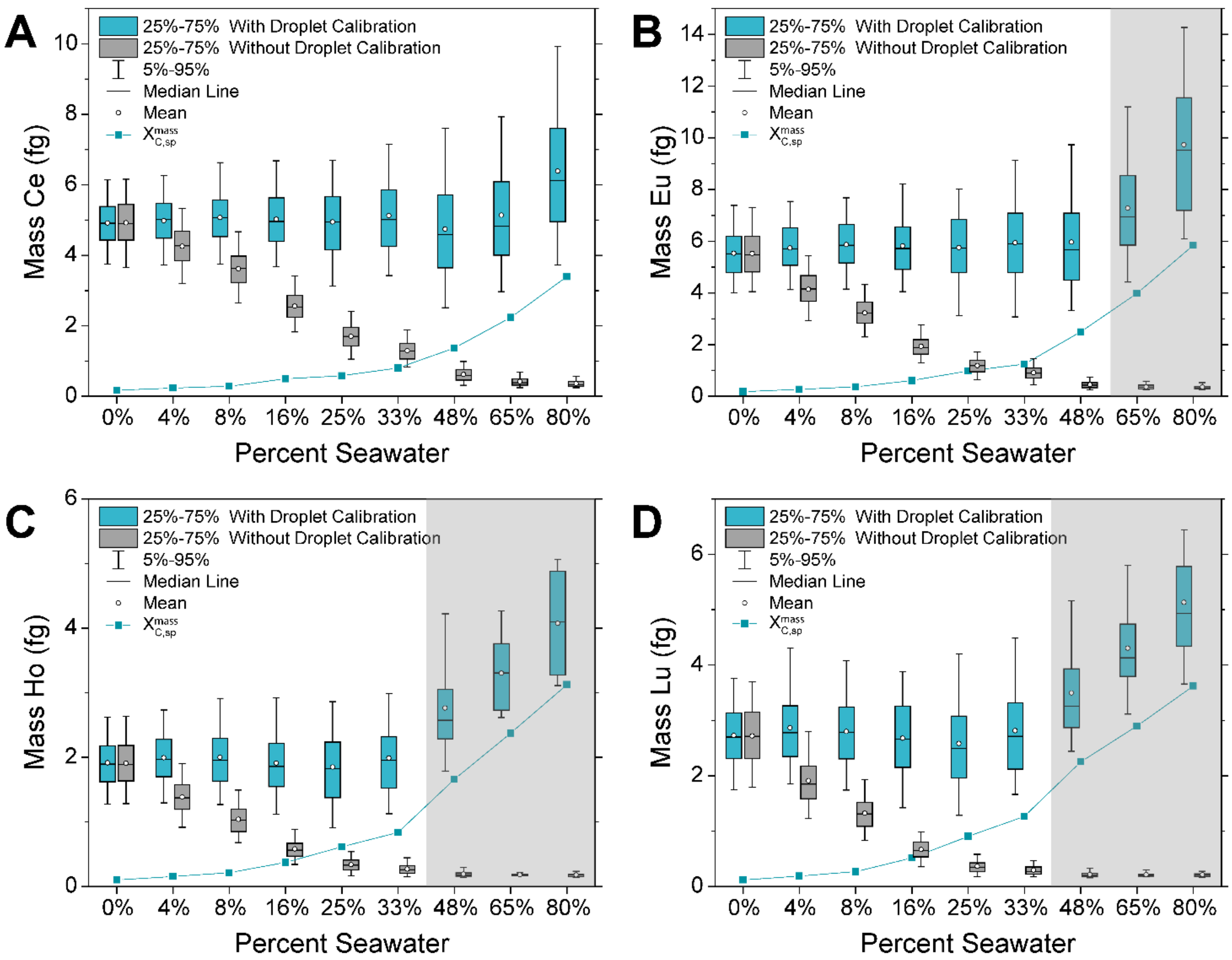

Figure 4. Box plots showing the mass distribution of REEs in the PS beads at different concentrations of seawater. Plots in which the $X_{C,sp}^{mass}$ exceeds the first quartile of the mass distributions measured with 0% seawater are shaded in gray. (A) Ce, (B) Eu, (C) Ho, (D) Lu.

Titanium Dioxide Particles in Seawater

Titanium dioxide nanoparticles (TiO_2 NPs) are a common ingredient in many mineral sunscreens. These NPs can then be released into the environment through recreational activity.[22, 51-53] TiO_2 NPs have been shown to have adverse effects on marine organisms.[54-55] In addition, sub-micron TiO_2, such as those used for pigments and food additives are predicted to be a major source of TiO_2 NPs in the environment.[56-58] Here, we measured polydisperse TiO_2 sub-micron particles in seawater to demonstrate the applicability of spICP-TOFMS for the measurement of particle pollutants in marine environments at environmentally relevant concentrations. In Figure 5, we report the measured mass distributions of Ti in the TiO_2 particles from 0% to 100% seawater concentrations with and without online microdroplet calibration. Without online microdroplet calibration, the median mass recovery of Ti in the TiO_2 particles decreases to 20% in the pure seawater matrix. With online microdroplet calibration, the median mass recovery is 119% in seawater. Moreover, up to a seawater concentration of 40% (v/v), the median mass measured with online microdroplet calibration is within 10% of the median mass measured in pure water. Importantly, the median mass recorded for the TiO_2 particles deviates from the true median at higher seawater concentrations because, as the $X_{C,sp,Ti}^{mass}$ increases, smaller TiO_2 particles (i.e. those with less Ti mass) are no longer recorded and this skews the mass distribution. In Figure S7, we provide the $X_{C,sp,Ti}^{mass}$ as a function of seawater concentrations, and in Figure S8 we provide Ti-mass histograms as a function of seawater concentration. These figures clearly show that the low-mass Ti signals are truncated with increasing seawater concentration, while the mass of Ti in the larger TiO_2 particles remains accurately determined.

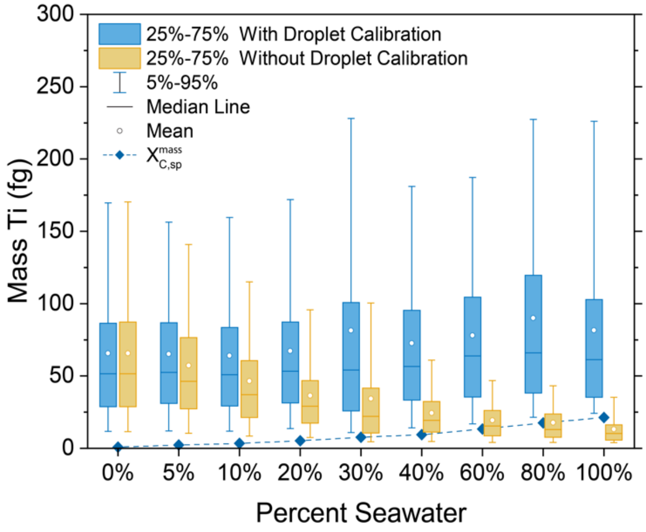

Figure 5. Mass distributions of E171 TiO_2 particles measured both with (blue) and without (yellow) online microdroplet calibration.

Conclusion

Online microdroplet calibration is a useful and high-throughput approach for the measurement of metal nanoparticles, microplastics, and metal-oxide sub-micron particles in seawater matrices. With online microdroplet calibration, microdroplet standards and analyte particles experience the same plasma conditions, and the approach can be used to correct for extreme matrix effects, such as those induced by high-salt-content samples like seawater. This approach can be used with a range of elements in the microdroplet standards and is well-suited for multiplexed analysis of diverse particle types in a single measurement. With online microdroplet calibration, we demonstrate accurate mass determinations of particles with up to 100-fold sensitivity attenuation. Since the method is automatically matrix-matched for each sample, it does not require onerous extraction methods or individualized matrix-matched standard preparation (which may not be possible with an unknown matrix composition) and thus improves analysis throughput and allows for more versatility in adjusting sample parameters such as dilution factors. Combined with spICP-TOFMS, this method is well-suited for applications such as the measurement of metals adsorbed to microplastic pollution in different environmental waters.

The potential implications of this research extend beyond the analysis of particles in seawater. Combined with spICP-TOFMS, this technique could be used to study adsorbed metals or metal additives in polymers, to characterize heterogeneous particles in variable solvents, or to measure particles in biological samples. Modern high-sensitivity ICP-MS instruments now enable the detection of carbon at the single-cell level, and this method could potentially be used to study endogenous elements in cells in environmental or biological matrices.

Supporting Information

Operating parameters and details of parameters used for microdroplet calibration. TEM images of Ag and Au NPs. Critical masses and diameters for all particles. Comparison of microdroplet signal to Ag and Au NP signal. Absolute sensitivities for ^{12}C and REEs in seawater. Histograms of TiO_2 NP sizes measured with and without microdroplet calibration in varying concentrations of seawater.

Acknowledgments

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References

1. Timerbaev, A. R.; Kuznetsova, O. V.; Keppler, B. K., Current trends and challenges in analysis and characterization of engineered nanoparticles in seawater. *Talanta* **2021,** *226*, 122201.
2. Auta, H. S.; Emenike, C. U.; Fauziah, S. H., Distribution and importance of microplastics in the marine environment: A review of the sources, fate, effects, and potential solutions. *Environment International* **2017,** *102*, 165-176.
3. Eriksen, M.; Lebreton, L. C. M.; Carson, H. S.; Thiel, M.; Moore, C. J.; Borerro, J. C.; Galgani, F.; Ryan, P. G.; Reisser, J., Plastic Pollution in the World's Oceans: More than 5 Trillion Plastic Pieces Weighing over 250,000 Tons Afloat at Sea. *PLOS ONE* **2014,** *9* (12), e111913.
4. Turner, A.; Holmes, L. A., Adsorption of trace metals by microplastic pellets in fresh water. *Environmental Chemistry* **2015,** *12* (5), 600-610.
5. Davranche, M.; Veclin, C.; Pierson-Wickmann, A.-C.; El Hadri, H.; Grassl, B.; Rowenczyk, L.; Dia, A.; Ter Halle, A.; Blancho, F.; Reynaud, S.; Gigault, J., Are nanoplastics able to bind significant amount of metals? The lead example. *Environ. Pollut.* **2019,** *249*, 940-948.
6. Prunier, J.; Maurice, L.; Perez, E.; Gigault, J.; Pierson Wickmann, A.-C.; Davranche, M.; Halle, A. t., Trace metals in polyethylene debris from the North Atlantic subtropical gyre. *Environ. Pollut.* **2019,** *245*, 371-379.
7. Ferreira, P.; Fonte, E.; Soares, M. E.; Carvalho, F.; Guilhermino, L., Effects of multi-stressors on juveniles of the marine fish Pomatoschistus microps: Gold nanoparticles, microplastics and temperature. *Aquat. Toxicol.* **2016,** *170*, 89-103.
8. Mos, B.; Kaposi, K. L.; Rose, A. L.; Kelaher, B.; Dworjanyn, S. A., Moderate ocean warming mitigates, but more extreme warming exacerbates the impacts of zinc from engineered nanoparticles on a marine larva. *Environ. Pollut.* **2017,** *228*, 190-200.
9. Oukarroum, A.; Polchtchikov, S.; Perreault, F.; Popovic, R., Temperature influence on silver nanoparticles inhibitory effect on photosystem II photochemistry in two green algae, Chlorella vulgaris and Dunaliella tertiolecta. *Environmental Science and Pollution Research* **2012,** *19* (5), 1755-1762.
10. Gniadek, M.; Dąbrowska, A., The marine nano- and microplastics characterisation by SEM-EDX: The potential of the method in comparison with various physical and chemical approaches. *Marine Pollution Bulletin* **2019,** *148*, 210-216.
11. Gondikas, A. P.; von der Kammer, F.; Reed, R. B.; Wagner, S.; Ranville, J. F.; Hofmann, T., Release of TiO2 nanoparticles from sunscreens into surface waters: a one-year survey at the old Danube recreational Lake. *Environ Sci Technol* **2014,** *48* (10), 5415-5422.
12. Lee, W.-W.; Chan, W.-T., Calibration of single-particle inductively coupled plasma-mass spectrometry (SP-ICP-MS). *J. Anal. At. Spectrom.* **2015,** *30* (6), 1245-1254.
13. Olesik, J. W.; Gray, P. J., Considerations for measurement of individual nanoparticles or microparticles by ICP-MS: determination of the number of particles and the analyte mass in each particle. *J. Anal. At. Spectrom.* **2012,** *27* (7), 1143-1155.
14. Kocic, J.; Günther, D.; Hattendorf, B., Improving detection capability for single particle inductively coupled plasma mass spectrometry with microdroplet sample introduction. *J. Anal. At. Spectrom.* **2021,** *36* (1), 233-242.

15. Montaño, M. D.; Badiei, H. R.; Bazargan, S.; Ranville, J. F., Improvements in the detection and characterization of engineered nanoparticles using spICP-MS with microsecond dwell times. *Environ. Sci.: Nano* **2014,** *1* (4), 338-346.
16. Ding, K.; Liang, S.; Xie, C.; Wan, Q.; Jin, C.; Wang, S.; Tang, Y. T.; Zhang, M.; Qiu, R., Discrimination and Quantification of Soil Nanoparticles by Dual-Analyte Single Particle ICP-QMS. *Anal Chem* **2022,** *94* (30), 10745-10753.
17. Borovinskaya, O.; Hattendorf, B.; Tanner, M.; Gschwind, S.; Günther, D., A prototype of a new inductively coupled plasma time-of-flight mass spectrometer providing temporally resolved, multi-element detection of short signals generated by single particles and droplets. *J. Anal. At. Spectrom.* **2013,** *28* (2), 226-233.
18. Borovinskaya, O.; Gschwind, S.; Hattendorf, B.; Tanner, M.; Gunther, D., Simultaneous mass quantification of nanoparticles of different composition in a mixture by microdroplet generator-ICPTOFMS. *Anal. Chem.* **2014,** *86* (16), 8142-8.
19. Hendriks, L.; Gundlach-Graham, A.; Hattendorf, B.; Günther, D., Characterization of a new ICP-TOFMS instrument with continuous and discrete introduction of solutions. *J. Anal. At. Spectrom.* **2017,** *32* (3), 548-561.
20. Mitrano, D. M.; Ranville, J. F.; Bednar, A.; Kazor, K.; Hering, A. S.; Higgins, C. P., Tracking dissolution of silver nanoparticles at environmentally relevant concentrations in laboratory, natural, and processed waters using single particle ICP-MS (spICP-MS). *Environmental Science: Nano* **2014,** *1* (3), 248-259.
21. Yang, Y.; Long, C.-L.; Li, H.-P.; Wang, Q.; Yang, Z.-G., Analysis of silver and gold nanoparticles in environmental water using single particle-inductively coupled plasma-mass spectrometry. *Sci. Total Environ.* **2016,** *563-564*, 996-1007.
22. Venkatesan, A. K.; Reed, R. B.; Lee, S.; Bi, X.; Hanigan, D.; Yang, Y.; Ranville, J. F.; Herckes, P.; Westerhoff, P., Detection and Sizing of Ti-Containing Particles in Recreational Waters Using Single Particle ICP-MS. *Bulletin of Environmental Contamination and Toxicology* **2018,** *100* (1), 120-126.
23. Phalyvong, K.; Sivry, Y.; Pauwels, H.; Gélabert, A.; Tharaud, M.; Wille, G.; Bourrat, X.; Ranville, J. F.; Benedetti, M. F., Assessing CeO2 and TiO2 Nanoparticle Concentrations in the Seine River and Its Tributaries Near Paris. *Frontiers in Environmental Science* **2021,** *8*.
24. Newman, K.; Metcalfe, C.; Martin, J.; Hintelmann, H.; Shaw, P.; Donard, A., Improved single particle ICP-MS characterization of silver nanoparticles at environmentally relevant concentrations. *J. Anal. At. Spectrom.* **2016,** *31* (10), 2069-2077.
25. Tan, S. H.; Horlick, G., Matrix-effect observations in inductively coupled plasma mass spectrometry. *J. Anal. At. Spectrom.* **1987,** *2* (8), 745-763.
26. Vanhaecke, F.; Dams, R.; Vandecasteele, C., 'Zone model' as an explanation for signal behaviour and non-spectral interferences in inductively coupled plasma mass spectrometry. *J. Anal. At. Spectrom.* **1993,** *8* (3), 433-438.
27. Agatemor, C.; Beauchemin, D., Matrix effects in inductively coupled plasma mass spectrometry: a review. *Anal. Chim. Acta* **2011,** *706* (1), 66-83.
28. Olesik, J. W.; Jiao, S., Matrix effects using an ICP-MS with a single positive ion lens and grounded stop: analyte mass dependent? *J. Anal. At. Spectrom.* **2017,** *32* (5), 951-966.
29. Olivares, J. A.; Houk, R. S., Suppression of analyte signal by various concomitant salts in inductively coupled plasma mass spectrometry. *Anal. Chem.* **1986,** *58* (1), 20-25.

30. Wimmer, A.; Urstoeger, A.; Funck, N. C.; Adler, F. P.; Lenz, L.; Doeblinger, M.; Schuster, M., What happens to silver-based nanoparticles if they meet seawater? *Water Res.* **2020,** *171*, 115399.
31. Toncelli, C.; Mylona, K.; Tsapakis, M.; Pergantis, S. A., Flow injection with on-line dilution and single particle inductively coupled plasma – mass spectrometry for monitoring silver nanoparticles in seawater and in marine microorganisms. *J. Anal. At. Spectrom.* **2016,** *31* (7), 1430-1439.
32. Pace, H. E.; Rogers, N. J.; Jarolimek, C.; Coleman, V. A.; Higgins, C. P.; Ranville, J. F., Determining transport efficiency for the purpose of counting and sizing nanoparticles via single particle inductively coupled plasma mass spectrometry. *Anal. Chem.* **2011,** *83* (24), 9361-9.
33. Montaño, M. D.; Olesik, J. W.; Barber, A. G.; Challis, K.; Ranville, J. F., Single Particle ICP-MS: Advances toward routine analysis of nanomaterials. *Anal. Bioanal. Chem.* **2016,** *408* (19), 5053-5074.
34. Huang, Y.; Tsz-Shan Lum, J.; Sze-Yin Leung, K., Single particle ICP-MS combined with internal standardization for accurate characterization of polydisperse nanoparticles in complex matrices. *J. Anal. At. Spectrom.* **2020,** *35* (10), 2148-2155.
35. Torregrosa, D.; Gómez-Pertusa, C.; Grindlay, G.; Gras, L.; Mora, J., Organics non-spectral interferences on nanoparticle characterization by means single particle inductively coupled plasma mass spectrometry. *J. Anal. At. Spectrom.* **2023**.
36. Aramendia, M.; Garcia-Mesa, J. C.; Alonso, E. V.; Garde, R.; Bazo, A.; Resano, J.; Resano, M., A novel approach for adapting the standard addition method to single particle-ICP-MS for the accurate determination of NP size and number concentration in complex matrices. *Anal. Chim. Acta* **2022,** *1205*, 339738.
37. Telgmann, L.; Metcalfe, C. D.; Hintelmann, H., Rapid size characterization of silver nanoparticles by single particle ICP-MS and isotope dilution. *J. Anal. At. Spectrom.* **2014,** *29* (7), 1265-1272.
38. Sötebier, C. A.; Kutscher, D. J.; Rottmann, L.; Jakubowski, N.; Panne, U.; Bettmer, J., Combination of single particle ICP-QMS and isotope dilution analysis for the determination of size, particle number and number size distribution of silver nanoparticles. *J. Anal. At. Spectrom.* **2016,** *31* (10), 2045-2052.
39. Gruszka, J.; Malejko, J.; Bajguz, A.; Godlewska-Żyłkiewicz, B., Method development for speciation analysis of silver nanoparticles and silver ions in green algae and surface waters at environmentally relevant concentrations using single particle ICP-MS. *J. Anal. At. Spectrom.* **2022,** *37* (6), 1208-1222.
40. Hendriks, L.; Ramkorun-Schmidt, B.; Gundlach-Graham, A.; Koch, J.; Grass, R. N.; Jakubowski, N.; Günther, D., Single-particle ICP-MS with online microdroplet calibration: toward matrix independent nanoparticle sizing. *J. Anal. At. Spectrom.* **2019,** *34* (4), 716-728.
41. Mehrabi, K.; Günther, D.; Gundlach-Graham, A., Single-particle ICP-TOFMS with online microdroplet calibration for the simultaneous quantification of diverse nanoparticles in complex matrices. *Environ. Sci.: Nano* **2019,** *6* (11), 3349-3358.

42. Harycki, S.; Gundlach-Graham, A., Online microdroplet calibration for accurate nanoparticle quantification in organic matrices. *Anal Bioanal Chem* **2022,** *414* (25), 7543-7551.
43. Gundlach-Graham, A.; Lancaster, R., Mass-Dependent Critical Value Expressions for Particle Finding in Single-Particle ICP-TOFMS. *Anal. Chem.* **2023,** *95* (13), 5618-5626.
44. Gundlach-Graham, A.; Hendriks, L.; Mehrabi, K.; Gunther, D., Monte Carlo Simulation of Low-Count Signals in Time-of-Flight Mass Spectrometry and Its Application to Single-Particle Detection. *Anal. Chem.* **2018,** *90* (20), 11847-11855.
45. Gondikas, A.; Gallego-Urrea, J.; Halbach, M.; Derrien, N.; Hassellöv, M., Nanomaterial Fate in Seawater: A Rapid Sink or Intermittent Stabilization? *Frontiers in Environmental Science* **2020,** *8*.
46. Bolea-Fernandez, E.; Rua-Ibarz, A.; Velimirovic, M.; Tirez, K.; Vanhaecke, F., Detection of microplastics using inductively coupled plasma-mass spectrometry (ICP-MS) operated in single-event mode. *J. Anal. At. Spectrom.* **2020,** *35* (3), 455-460.
47. Gonzalez de Vega, R.; Goyen, S.; Lockwood, T. E.; Doble, P. A.; Camp, E. F.; Clases, D., Characterisation of microplastics and unicellular algae in seawater by targeting carbon via single particle and single cell ICP-MS. *Anal. Chim. Acta* **2021,** *1174*, 338737.
48. Laborda, F.; Trujillo, C.; Lobinski, R., Analysis of microplastics in consumer products by single particle-inductively coupled plasma mass spectrometry using the carbon-13 isotope. *Talanta* **2021,** *221*, 121486.
49. Hendriks, L.; Mitrano, D. M., Direct Measurement of Microplastics by Carbon Detection via Single Particle ICP-TOFMS in Complex Aqueous Suspensions. *Environmental Science & Technology* **2023,** *57* (18), 7263-7272.
50. Harycki, S.; Gundlach-Graham, A., Characterization of a high-sensitivity ICP-TOFMS instrument for microdroplet, nanoparticle, and microplastic analyses. *J. Anal. At. Spectrom.* **2023,** *38* (1), 111-120.
51. Gondikas, A.; von der Kammer, F.; Kaegi, R.; Borovinskaya, O.; Neubauer, E.; Navratilova, J.; Praetorius, A.; Cornelis, G.; Hofmann, T., Where is the nano? Analytical approaches for the detection and quantification of TiO2 engineered nanoparticles in surface waters. *Environ. Sci.: Nano* **2018,** *5* (2), 313-326.
52. Slomberg, D. L.; Auffan, M.; Guéniche, N.; Angeletti, B.; Campos, A.; Borschneck, D.; Aguerre-Chariol, O.; Rose, J., Anthropogenic Release and Distribution of Titanium Dioxide Particles in a River Downstream of a Nanomaterial Manufacturer Industrial Site. *Frontiers in Environmental Science* **2020,** *8*.
53. Heilgeist, S.; Sahin, O.; Sekine, R.; Stewart, R. A., Mapping the Complex Journey of Swimming Pool Contaminants: A Multi-Method Systems Approach. *Water* **2022,** *14* (13), 2062.
54. Barmo, C.; Ciacci, C.; Canonico, B.; Fabbri, R.; Cortese, K.; Balbi, T.; Marcomini, A.; Pojana, G.; Gallo, G.; Canesi, L., In vivo effects of n-TiO2 on digestive gland and immune function of the marine bivalve Mytilus galloprovincialis. *Aquat. Toxicol.* **2013,** *132-133*, 9-18.
55. Moore, M. N., Do nanoparticles present ecotoxicological risks for the health of the aquatic environment? *Environment International* **2006,** *32* (8), 967-976.

56. Zheng, Y.; Nowack, B., Size-Specific, Dynamic, Probabilistic Material Flow Analysis of Titanium Dioxide Releases into the Environment. *Environ. Sci. Technol.* **2021,** *55* (4), 2392-2402.
57. Kaegi, R.; Ulrich, A.; Sinnet, B.; Vonbank, R.; Wichser, A.; Zuleeg, S.; Simmler, H.; Brunner, S.; Vonmont, H.; Burkhardt, M.; Boller, M., Synthetic TiO2 nanoparticle emission from exterior facades into the aquatic environment. *Environ. Pollut.* **2008,** *156* (2), 233-239.
58. Loosli, F.; Wang, J.; Rothenberg, S.; Bizimis, M.; Winkler, C.; Borovinskaya, O.; Flamigni, L.; Baalousha, M., Sewage spills are a major source of titanium dioxide engineered (nano)-particle release into the environment. *Environ. Sci.: Nano* **2019,** *6* (3), 763-777.