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Nanoparticles in drinking water

Synthetic nanoparticles are increasingly being used in everyday products, but little is known about environmental releases of these materials. Our knowledge of how many natural nanoparticles occur in drinking water is also inadequate. An initial assessment is presented here.

Drinking water is clear, and yet it contains millions of particles. Substances are described as particulate – as opposed to dissolved – if they are retained by a filter with a pore diameter of 0.45 µm. However, this operational definition overlooks the fact that water contains many smaller particles which can pass through such filters. These include particles measuring 1–100 nm, i. e. nanoparticles. It is not known how many of these naturally occurring nanoparticles are actually present in drinking water. Our aim, therefore, was to make an initial assessment of natural nanoparticles in drinking water, using various methods of microscopy.

On this basis, it should in future be possible to distinguish between natural and man-made nanoparticles. The latter are now widely used in consumer products, e. g. in sunscreens (titanium dioxide), textiles (silver) and cosmetics (fullerene), as well as in facades (photocatalytic titanium dioxide) and scratch-proof paints (silicon dioxide). However, little is known about releases or the fate of synthetic nanoparticles in the environment.

Samples prepared by sedimentation/centrifugation. Based on the method described by Perret [1], we developed a technique for the analysis of nanoparticles. After samples have been collected – we used drinking water from the Lengg waterworks in Zurich – the nanoparticles first need to be separated from the larger particles. The simplest and quickest way of achieving this is by filtration. In principle, small particles pass through the pores while larger ones are retained. During the filtration process, however, the filter pores become blocked, leading to the retention of ever-smaller particles. To avoid this problem, we opted for stepwise sedimentation/centrifugation. In this procedure, the water sample is first allowed to stand for 2 hours in a 30-litre sedimentation tank. During this time, the large particles settle more rapidly than the small ones and can thus be separated out. Next, the uppermost 2 cm of water (= 1 litre) is carefully drawn off with a peristaltic pump (see photo). However, many of the particles in this water fraction are still larger than 100 nm. To allow these to be also separated from the nanoparticles, the sample is then centrifuged for 30 minutes at 330 *g* (*g* = acceleration due to gravity). The uppermost layer (2 cm) is once again removed with a peristaltic pump and undergoes a second centrifugation step (1 hour at 2700 *g*). The resulting supernatant contains mainly nanoparticles.

The nanoparticles now have to be mounted in such a way that they can be viewed under the microscope. This is done with the aid of ultracentrifugation (12 hours at 120 000 *g*). The fractionation steps, including the various particle diameter cut-offs, are summarized in the Table. As the cut-off diameter also depends on particle density, two different densities are given in each case.

Fractionation steps used for sample preparation.

Fractionation method	Time	Force applied (*g*)	Cut-off (1.1 g/cm^3)	Cut-off (2 g/cm^3)
Sedimentation	2 h	1 × *g*	9 µm	3 µm
Centrifugation 1	0.5 h	330 × *g*	750 nm	250 nm
Centrifugation 2	1 h	2700 × *g*	180 nm	60 nm
Ultracentrifugation	12 h	120 000 × *g*	12 nm	4 nm

Brian Sinnet preparing a sample at the Lengg lake water treatment plant (Zurich).

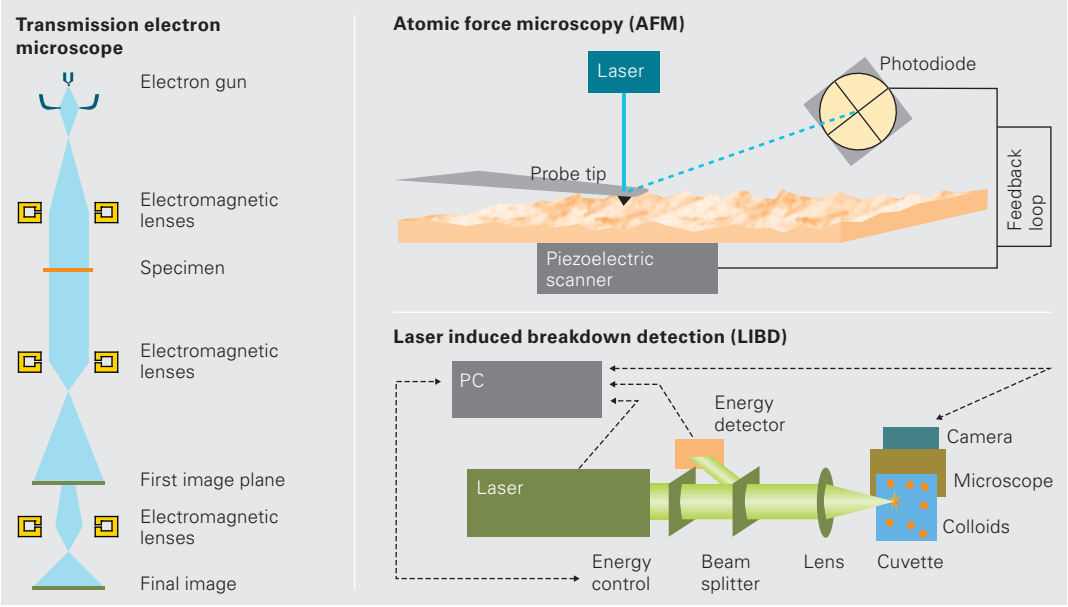


Fig. 1: Schematic view of the microscopy methods used (LIBD simplified from [3]).

The 1.1 g/cm^3 density is roughly equivalent to that of organic particles, and 2 g/cm^3 to that of clay minerals.

Particle surfaces scanned in the atomic force microscope. To analyse the nanoparticles, we used various methods of microscopy. Each method provides specific information on certain properties of the particles, but also generates artefacts. However, by comparing the results derived from the various analytical procedures, it is possible to identify artefacts and produce a comprehensive characterization of the particles.

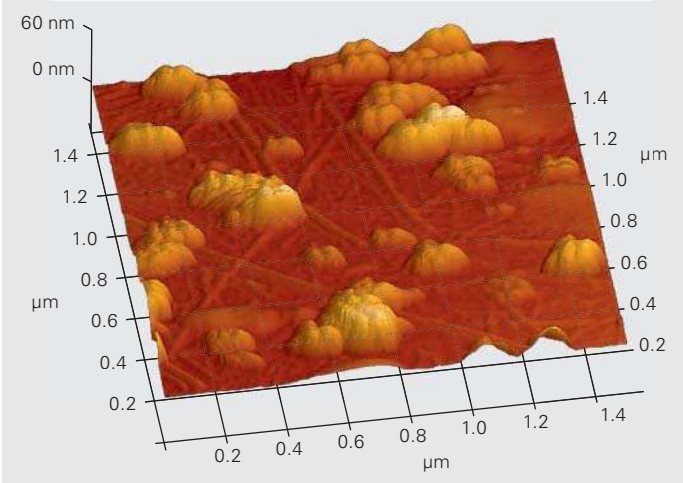
In atomic force microscopy (AFM), the surface of the particles is scanned with a very fine tip (Fig. 1). In the process, the probe tip is brought so close to the specimen that Van der Waals forces – relatively weak non-covalent interactions between atoms or molecules – become detectable. Using this technique, the topography of the sample can be mapped with a high degree of precision. However, the particles have to be placed on an extremely flat specimen holder. Freshly cleaved sheets of mica (a naturally occurring sheet silicate) are generally used for this purpose.

On the basis of the AFM findings and descriptions in the literature, two types of particles can be distinguished: spherical aggregates, probably humic acid aggregates, with a diameter (height) of up to 60 nm and fibrous particles, presumably polysaccharides, which are several hundred nanometres long and only a few nanometres high (Fig. 2). These results accord well with those obtained by Santschi, who studied marine particles [2].

Particle size determined in the transmission electron microscope. In transmission electron microscopy (TEM), a specimen is illuminated by a beam of electrons (Fig. 1). While the majority will pass unimpeded through the sample, electrons will be deflected when they hit matter, such as a particles deposited on the sample carrier. The electrons transmitted through the sample are detected by a camera, forming the TEM image. Wherever a particle is located, the electrons are more strongly scattered, which translates into dark areas in the image. For TEM analysis, therefore, particles have to be mounted on a specimen holder that permits the passage of electrons. A copper grid coated with a thin film of carbon proved to be most suitable for our applications.

Figure 3 shows nanoparticles in drinking water imaged with a transmission electron microscope. The contrast was enhanced by strongly defocusing the electron beam, producing an image which is in principle of poor quality, but highly suitable for image analysis. In purely qualitative terms, the same types of particles are visible here as in the AFM analysis. With the aid of image

Fig. 2: Nanoparticles in drinking water, visualized by atomic force microscopy (AFM).



analysis tools, the particles can be detected and measured automatically. For our project, we studied 1800 particles on a total of 10 TEM images. On the basis of these data, the size distribution of nanoparticles in drinking water can be determined. With the TEM settings used, it is possible to measure particles down to a minimum diameter of 40 nm. The results indicate that the particle size distribution follows a power law. When the values are plotted on a log-log scale, the particle size distribution lies on a straight line with a slope of –3.3. This accords well with textbook accounts of particle size distribution in natural waters [4].

Particle concentration and average size readily determined by laser-induced breakdown detection. The microscopy techniques applied to date provide detailed information on individual particles. In combination with digital image processing, large numbers of individual particles can be measured. However, this is extremely time-consuming, as the samples first have to be prepared and then manually analysed under the microscope. We therefore tested a new laser-based technique known as laser-induced breakdown detection (LIBD), which makes it possible to obtain information on the mean particle diameter and the total number of particles within a few minutes. Our measurements were carried out on a prototype LIBD system at the Research Center in Karlsruhe, Germany.

A schematic view of how the LIBD operates is given in Fig. 1. A 20-Hz pulsed laser beam (green, 532 nm) is focused into a measurement cuvette. Whenever a particle lies within the laser focus, it breaks down, and generates a plasma (an ionized gas containing free charge carriers such as ions or electrons). This emission of light in the visible spectrum is recorded by a specialised camera. The position of the particle on the image (x/y coordinates) together with the frequency (ratio of laser pulses delivered to plasma events detected) is used to determine the mean diameter and the concentration of particles in the sample. For our sample, it was shown that a litre of drinking water contains approx. 7×10^{11} particles. The mean particle diameter is around 15 nm.

Comparing the various techniques. Overall, the results we obtained with the three different microscopy methods show a good measure of agreement. With both AFM and TEM, two different particle types were identified – spherical aggregates (humic acid aggregates) and elongated, fibrous particles (polysaccharides). On the basis of several TEM images, the particle size distribution was calculated. For particles > 40 nm, the size distribution follows a power law. In order to compare the results of the TEM (particles > 40 nm) and LIBD (particles > 10 nm) measurements, we had to extrapolate the size distribution determined experimentally by TEM down to 10 nm. The total concentration of particles > 10 nm would then be around 1.1×10^{12} particles per litre, with a mean diameter of 13 nm, which is in close agreement with the results of the LIBD measurements (7×10^{11} particles per litre, 15 nm).

Needles in a haystack. We used various analytical methods to study the nanoparticles that occur naturally in drinking water.

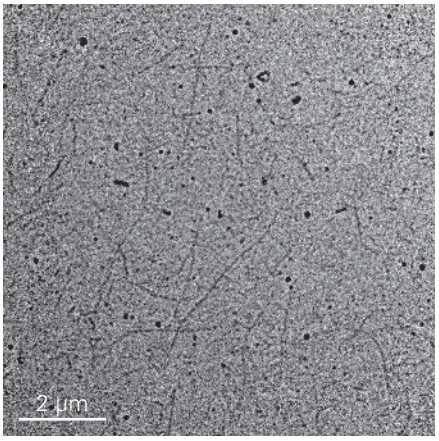


Fig. 3: Transmission electron micrograph of nanoparticles in drinking water.

A precise characterization of the natural nanoparticles already present is required in order to distinguish them from synthetic nanoparticles that could enter the water cycle in the future. As yet, however, little is known about the release and environmental behaviour of synthetic nanoparticles, or about the quantities to be expected in drinking water or untreated water. In addition, according to our findings, drinking water already contains vast quantities of natural nanoparticles; the detection of synthetic particles will therefore pose the next major challenge for researchers. ○ ○ ○

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