

Effects of engineered nanoparticles

As nanoparticles are already widely used, it was only too likely that they would ultimately find their way into the environment. This has now been demonstrated for the first time by Eawag studies – which also show how engineered nanoparticles can exert toxic effects on aquatic organisms.

Nanoparticles are found in increasing numbers of products, making it all the more likely that they will also be released into the aquatic environment. This may occur in the course of production, use or disposal, either directly (e.g. as a result of accidents) or via wastewater. Although the proportion of engineered nanoparticles in the environment remains low, compared with natural particles [1], there is still a need to assess the environmental risks. This calls not only for analytical systems to permit quantification but also for information on input pathways, amounts, and the fate and toxicity of engineered nanoparticles in natural waters. Initial studies describing the adverse effects of nanoparticles on aquatic organisms have now been published; overall, however, knowledge in this area is largely lacking. The situation is complicated by the existence of a very broad spectrum of engineered nanoparticles, differing in their chemical, physical and morphological properties (see Box “Engineered nanoparticles”). But precisely these properties need to be taken into account in efforts to understand how nanoparticles act on organisms, since they influence the bio-availability of particles and the mechanisms of toxicity. Eawag is therefore currently carrying out studies on the release and toxicity of engineered nanoparticles, and developing analytical methods for their characterization and detection in aquatic systems.

Titanium dioxide nanoparticles released from facades. It took almost 100 years for the detrimental effects of particles of a different type – asbestos fibres – to be finally recognized. Today, asbestos is banned in Switzerland, the EU and many other countries. To avoid a similar scenario arising with nanoparticles, it is essential to assess at an early stage the extent to which these particles are released. It is now generally agreed that nanoparticles do end up in the environment sooner or later, but this has not been specifically shown to date. However, such data are indispensable if we are to be able to predict future concentrations of nanoparticles in various environmental compartments (air, water, soil).

In an initial case study, the leaching of whitening pigments from facades was therefore investigated [3]. These engineered particles of titanium dioxide (TiO₂) are of various sizes, which can be described by a Gaussian distribution. On average, the TiO₂ particles measure about 150 nm (based on the total number), but approx. 10 % fall into the nanoparticle category (<100 nm). Thanks to their spherical shape, the whitening pigments can be readily distinguished from natural (geogenic) TiO₂ particles. In the study, facades, facade runoff and urban stormwater runoff (discharged into receiving waters) were analysed for engineered TiO₂ nanoparticles. Figure 1A shows a new facade, in which indi-



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Engineered nanoparticles

Engineered nanoparticles are nanotechnologically produced solid particles with a diameter of 100 nm or less (1 nanometre = a millionth of a millimetre), which take a wide variety of forms. They may consist of metals or metal oxides (inorganic, e.g. silver or titanium dioxide nanoparticles) or of carbon (organic nanoparticles). They can vary in shape (tubes, spheres, disks, fibres) and surface properties (untreated or chemically modified). Depending on the conditions, they can occur as individual particles or as aggregates in suspension. The artificial particles exhibit novel mechanical, electronic, chemical or optical properties, which are due to their high surface-to-volume ratio and make them ideally suited for various scientific, medical, industrial and commercial applications. Over 800 nanotech products are already available on the market. These include, for example, water-repellent textiles, ultraviolet-absorbing sunscreens, and antibacterial cosmetics, deodorants and toothpastes, as well as paints and varnishes that give surfaces a self-cleaning, antimicrobial or scratch-resistant coating. One of the most widely used nanomaterials is silver [2]. Often, however, it is not known whether products contain silver in the form of a salt (e.g. silver nitrate or silver chloride) or as engineered silver nanoparticles.

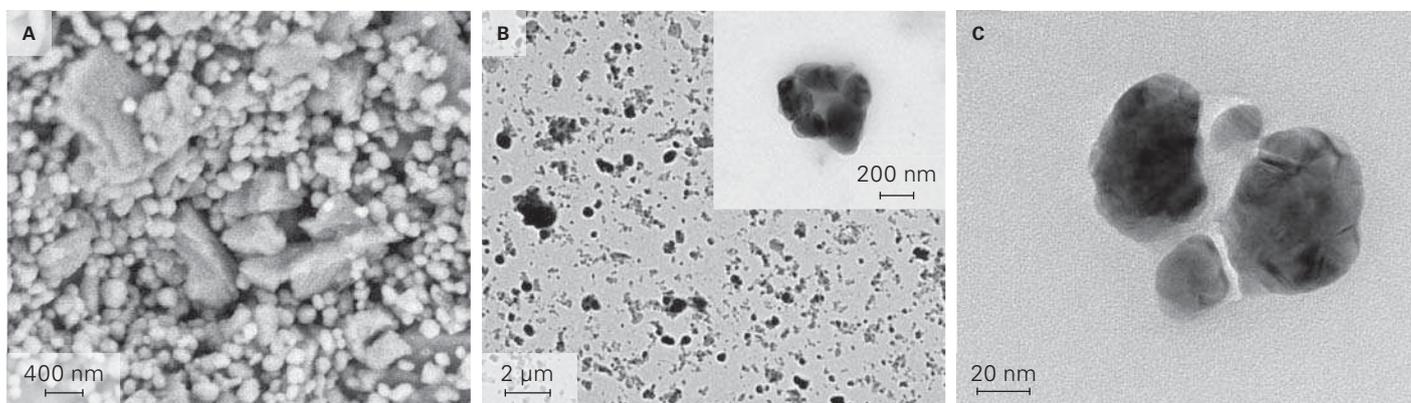


Fig. 1: Titanium dioxide nanoparticles in a facade (A), in facade runoff (B) and in urban stormwater runoff discharged into receiving waters (C).

vidual TiO₂ particles are clearly recognizable as bright spots. Very similar particles were also found in the facade runoff (Fig. 1B). Particles of the same size and shape were likewise detected in the urban runoff (Fig. 1C), which suggests that they mainly originate from the facades. From an (eco-)toxicological viewpoint, whitening pigments do not raise significant concerns, but they are well-suited as a model substance for evaluating the transport of nanoparticles from urban areas to the aquatic environment. It can be assumed that other nanoparticles will behave in a similar way to TiO₂ particles.

Silver nanoparticles from facades also found in water. Far more critical, in terms of ecotoxicology, is the case of engineered silver nanoparticles (see Box “Ecotoxicity of silver”), which are also used, for example, in facade paints. Engineered silver nanoparticles consist of metallic, uncharged silver atoms, although the particle surfaces may also be modified by inorganic or organic substances. As a result, they obtain a positive or negative charge, which prevents aggregation of the particles. The leaching of engineered silver nanoparticles from a facade was initially investigated in a simulation chamber, in which the facade was exposed to rain and sunlight under controlled conditions. Microscopic analysis of the facade runoff indicated that silver nanoparticles 5–10 nm in size are indeed leached from the facade. Alongside the chamber experiments, the same paint on a model house was exposed to natural weathering. Interestingly, the initial results show that silver nanoparticles are released from the model house in even larger quantities – even though the conditions in the simulation chamber are considerably harsher and much more water was used. It is thus clear that silver nanoparticles, like TiO₂ particles, can enter natural waters. But what are the effects of silver nanoparticles in the aquatic environment?

Toxicity of silver nanoparticles partly due to silver ions.

Another case study was carried out to investigate the effects of engineered silver nanoparticles on the photoengineered activity of the model green alga *Chlamydomonas reinhardtii* [4]. For this purpose, we used a suspension of metallic, carbonate-coated silver particles with an average size of 25 nm (Fig. 2). Because

the metallic silver particles are produced from silver ions (Ag⁺), a small residue of silver ions (in our case around 1%) is always present in suspensions of this kind. In our ecotoxicology experiments, we aimed to find out whether the particle suspension has toxic effects on photosynthesis and, if so, whether the toxicity is caused by the silver ions (see Box “Ecotoxicity of silver”) or by the nanoparticles themselves.

The algae were exposed to the suspension of silver nanoparticles for 1 or 2 hours. In addition, to compare the effects of the particles directly with those of silver ions, experiments were performed with a silver nitrate solution. It was found that algal photosynthesis is increasingly inhibited as total silver concentrations rise, and that silver nitrate is more toxic than the silver

Ecotoxicity of silver

Silver (Ag) has long been known to have antimicrobial effects: in the 19th century the metal was already used as an antibiotic, and it was recognized even earlier that drinking water keeps better in silver vessels. The use of silver as a disinfectant is based on the fact that it is toxic to a broad spectrum of bacteria and has relatively low toxicity to humans. For various aquatic organisms, silver is also among the most toxic metals. It is persistent and at relatively high concentrations in water, it can accumulate both in sediments and in organisms. As for other metals, the toxicity of silver is dependent on the bioavailability of silver ions (Ag⁺), which is influenced in turn by the chemical composition of the experimental medium, or water. The toxic effects of silver ions can be reduced by the presence of complexing agents, but at the same time the toxicity of silver is essentially based precisely on its strong affinity for sulfhydryl, amino and phosphate groups, which leads in organisms to the formation of complexes with various biomolecules.

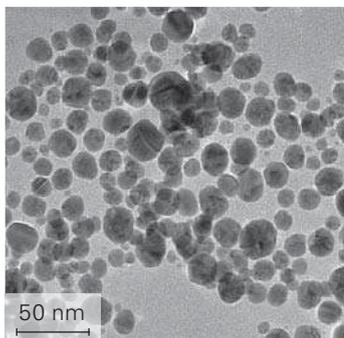


Fig. 2: Transmission electron micrograph of the engineered silver nanoparticles used in our study.

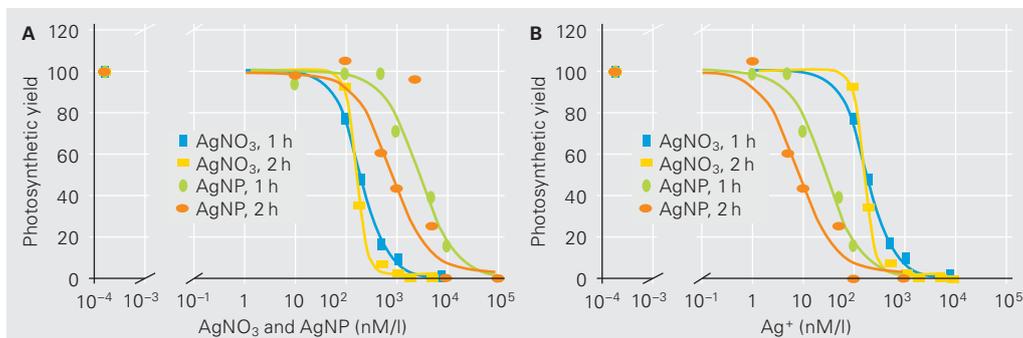


Fig. 3: Toxic effects of silver nanoparticles (NP) and a silver nitrate solution on photosynthetic activity in the green alga *Chlamydomonas reinhardtii* plotted as a function of the total (= metallic + ionic) silver concentration (A) and the silver ion concentration alone (B).

nanoparticles (Fig. 3A). To separate the effects of the particles from those of the silver ions, the experiments were repeated in the presence of cysteine. This amino acid forms strong complexes with silver ions, so that the ions are subsequently no longer available to the algae. In this case, the toxic effects of the silver nanoparticle suspension were completely abolished in the presence of an excess of cysteine – i. e. the toxicity of the particle suspension must be based on the effects of the silver ions.

Surprisingly, if the toxicity of the nanoparticles and silver nitrate is plotted as a function of the silver ion concentration (Fig. 3B), it becomes apparent that silver nanoparticles are more toxic than silver nitrate. However, this effect cannot be wholly explained by the silver ions present in the particle suspension, as the concentration is too low. Other processes must also be involved. One logical explanation is that, through contact with the algae,

the silver nanoparticles release larger quantities of silver ions. As we studied toxicity solely in terms of photosynthetic activity, we cannot exclude the possibility that nanoparticles also enter the algae, triggering additional toxic mechanisms. We are currently investigating this hypothesis, and provisional findings indicate that silver nanoparticles are indeed taken up by algal cells.

Environmental releases of nanoparticles to be avoided! Our findings confirm that nanoparticles are released into the environment, and that the associated ecotoxicological risks are not to be underestimated. This applies not only for aquatic but also for terrestrial systems. Further studies are required, also covering other nanomaterials. In addition, standard methods need to be developed for the detection of nanoparticles and for the assessment of toxicity (cf. the article on the Ecotox Centre on p. 38).

Manufacturers should commit themselves to sensible use of nanoparticles. It is also important to educate consumers – firstly, through consistent labelling (at present, the composition of products is frequently unclear) and specific instructions on handling, and also by informing consumer associations about potential health and environmental risks. The priority goal should be to prevent nanoparticles spreading in the environment. ○○○

Research under real conditions: the experimental house used to determine the release of nanoparticles from facades.



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