

Fate and Risks of Nanomaterials in Aquatic and Terrestrial Environments

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CONSPECTUS

Over the last decade, nanoparticles have been used more frequently in industrial applications and in consumer and medical products, and these applications of nanoparticles will likely continue to increase. Concerns about the environmental fate and effects of these materials have stimulated studies to predict environmental concentrations in air, water, and soils and to determine threshold concentrations for their ecotoxicological effects on aquatic or terrestrial biota.

Nanoparticles can be added to soils directly in fertilizers or plant protection products or indirectly through application to land or wastewater treatment products such as sludges or biosolids. Nanoparticles may enter aquatic systems directly through industrial discharges or from disposal of wastewater treatment effluents or indirectly through surface runoff from soils. Researchers have used laboratory experiments to begin to understand the effects of nanoparticles on waters and soils, and this Account reviews that research and the translation of those results to natural conditions.

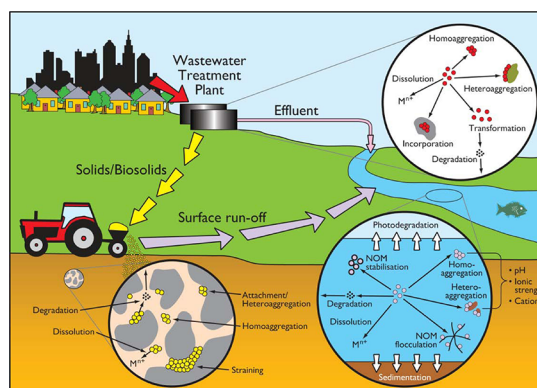
In the environment, nanoparticles can undergo a number of potential transformations that depend on the properties both of the nanoparticle and of the receiving medium. These transformations largely involve chemical and physical processes, but they can involve biodegradation of surface coatings used to stabilize many nanomaterial formulations.

The toxicity of nanomaterials to algae involves adsorption to cell surfaces and disruption to membrane transport. Higher organisms can directly ingest nanoparticles, and within the food web, both aquatic and terrestrial organisms can accumulate nanoparticles.

The dissolution of nanoparticles may release potentially toxic components into the environment. Aggregation with other nanoparticles (homoaggregation) or with natural mineral and organic colloids (heteroaggregation) will dramatically change their fate and potential toxicity in the environment. Soluble natural organic matter may interact with nanoparticles to change surface charge and mobility and affect the interactions of those nanoparticles with biota. Ultimately, aquatic nanomaterials accumulate in bottom sediments, facilitated in natural systems by heteroaggregation. Homoaggregates of nanoparticles sediment more slowly.

Nanomaterials from urban, medical, and industrial sources may undergo significant transformations during wastewater treatment processes. For example, sulfidation of silver nanoparticles in wastewater treatment systems converts most of the nanoparticles to silver sulfides (Ag₂S). Aggregation of the nanomaterials with other mineral and organic components of the wastewater often results in most of the nanomaterial being associated with other solids rather than remaining as dispersed nanosized suspensions.

Risk assessments for nanomaterial releases to the environment are still in their infancy, and reliable measurements of nanomaterials at environmental concentrations remain challenging. Predicted environmental concentrations based on current usage are low but are expected to increase as use increases. At this early stage, comparisons of estimated exposure data with known toxicity data indicate that the predicted environmental concentrations are orders of magnitude below those known to have environmental effects on biota. As more toxicity data are generated under environmentally-relevant conditions, risk assessments for nanomaterials will improve to produce accurate assessments that assure environmental safety.



Introduction

Since the manufactured nanomaterials industry began its growth less than a decade ago, concern has been raised about the possible entry of nanomaterials into aquatic and terrestrial environments, and their ultimate impact on biota. A key question for regulators is whether nanomaterials require different regulation from that for equivalent micrometer-sized materials, i.e. does nanosize result in greater bioavailability and toxicity. Here, the fate of nanomaterials entering the natural environment, especially changes in particle size, surface charge, and chemical form, will be critical controls of bioavailability.

Research undertaken in synthetic media under controlled laboratory conditions showed that the important processes controlling nanoparticle behavior were dissolution and aggregation.^{1–3} It was recognized, however, that, in natural aquatic and terrestrial environments, nanomaterial fate can differ significantly from that in synthetic media, largely because of the dominant presence of natural nanoparticles and colloidal materials.⁴ The study of these interactions represents the major challenge which, to date, has received little attention, largely because of the experimental difficulties in undertaking studies of particle size at environmentally relevant concentrations typically in the $\mu\text{g/L}$ or $\mu\text{g/kg}$ range.

This Account reviews the current status of experiments in synthetic waters/soils and how research is beginning to translate these to natural conditions.

Nanomaterials Definition

Nanoparticles occur naturally in aquatic and terrestrial environments, in the finer fractions of colloidal clays, mineral precipitates (aluminum, iron and manganese oxides, and hydroxides), and dissolved organic matter (humic and fulvic acids). In 2008, the International Organization for Standardization (ISO) classified nanomaterials into three main groups: nanoparticles (all three dimensions between 1 and 100 nm); nanoplates (one dimension between 1 and 100 nm); and nanofibers (two dimensions between 1 and 100 nm).⁵ Manufactured nanomaterials comprise seven main classes: carbonaceous nanomaterials (e.g., carbon nanotubes (CNTs)); semiconductors (e.g., quantum dots); metal oxides (e.g., zinc oxide); nanopolymers (e.g., dendrimers); nanoclays; emulsions (e.g., acrylic latex); and metals (e.g., silver). These nanomaterials may exist in single, aggregated, or agglomerated forms and have various shapes, coatings, and surface functionality. The regulation of nanomaterials remains a challenge for agencies applying existing frameworks designed to address the environmental safety of chemicals.

Nanomaterial Solubility

In nanomaterial studies, the term solubility has been loosely applied, especially in relation to carbon-based nanomaterials, often meaning “forming stabilized suspensions” rather than truly dissolving. Hydrophobic nanomaterials such as CNTs or fullerenes are virtually insoluble in natural waters (the solubility of fullerene has been calculated as 10^{-18} mol/L⁶), and dissolution requires extreme measures, such as solvent addition, although lengthy sonication appears to achieve a stable suspension. Surface functionalization is necessary to enhance dispersion, although such derivatives were less cytotoxic as a result of a reduced ability to generate reactive oxygen species.⁷

Most metal-based nanoparticles are hydrophilic and have a finite but often low solubility. In many studies, this is not measured, despite the soluble ionic metal fraction being the most toxic to aquatic and terrestrial biota. Franklin et al.,¹ investigating the biological impacts of ZnO nanoparticles, found that, despite a common belief that ZnO was “insoluble”, nanoparticulate ZnO rapidly dissolved to produce 6 mg/L of dissolved (dialyzable) Zn within 6 h and 16 mg/L in 72 h in a buffered pH 7.5 soft water, in excess of the 5 mg Zn/L that would be toxic to most aquatic biota. By contrast, nanoparticulate cerium oxide (CeO_2) has a very low solubility (ng/L), and so the effects of nanoparticle versus macroparticle toxicity could be readily investigated in the absence of the confounding process of dissolution.⁸ Greater toxicity to algae was observed for nanoparticulate CeO_2 compared to its macroparticulate equivalent (Figure 1).

Semiconductor quantum dots based on cadmium selenide (CdSe) have been shown to release ionic cadmium (Cd) as a result of selenide oxidation;⁹ however, experiments were conducted at unnaturally highly concentrated solutions (250 mg/L) when as high as 80 mg Cd/L was observed, greatly exceeding accepted water quality guidelines.¹⁰

Dissolution of silver (Ag) nanoparticles involves the oxidation of surface elemental Ag to Ag^+ and subsequent desorptive dissolution. This requires oxygen and protons, and it has been postulated that peroxide radicals are active intermediates,¹¹ but superoxide radicals were not detected. Nanoparticulate Ag was shown to be more toxic than micrometer-sized Ag (Angel et al., CSIRO, unpublished data), largely due to the greater release of ionic Ag (Figure 1).

The toxicity to algal species shown in Figure 1 involves adsorption to the cell surface and disruption to membrane transport. In higher organisms, direct ingestion of nanoparticles has been demonstrated (e.g. ref 12); however, trophic

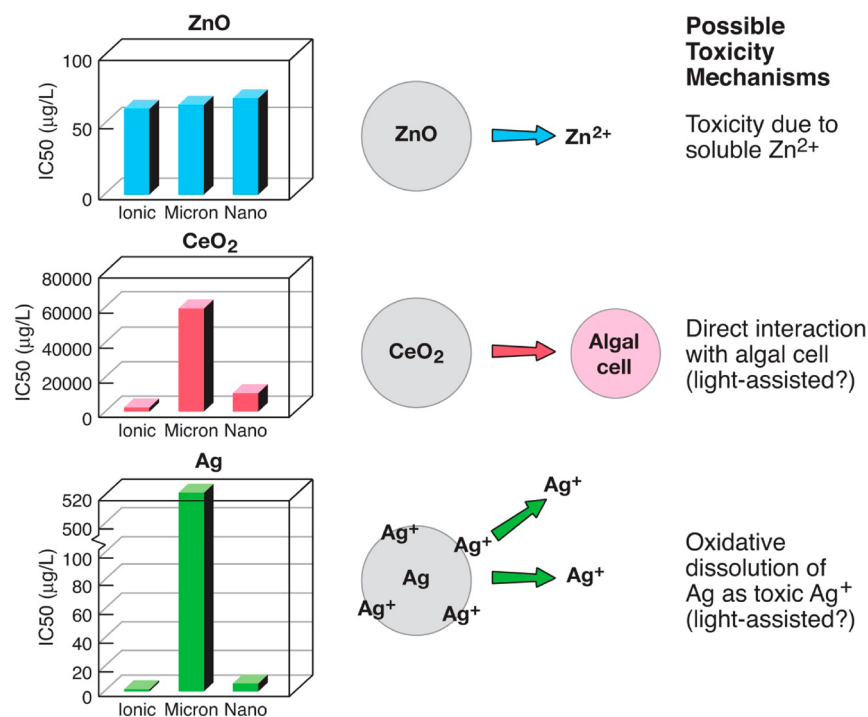


FIGURE 1. Comparison of the toxicities of micrometer and nanosized ZnO, CeO₂, and Ag and their ionic equivalents Zn²⁺, Ce³⁺, and Ag⁺ to the freshwater alga *Pseudokirchneriella subcapitata* in a synthetic soft water (toxicity values are IC₅₀s, the concentrations causing a 50% growth inhibition in µg/L) (respectively from refs 1, 8, and Angel et al., CSIRO, unpublished results).

transfer, e.g. via nanoparticle-coated algae, is a viable, yet so far poorly studied, uptake pathway.^{13–15} Such studies in natural waters (and soils) at environmental concentrations are needed.

Nanoparticle Aggregation

Aquatic and terrestrial environments contain a range of natural nanoparticles, including colloidal clays, iron and manganese hydrous oxides, and dissolved organic matter (comprising fulvic and humic acids), fibrillar colloids (exopolymers) that are exudates from algae, and other microorganisms (these are largely polysaccharides and some proteins).¹⁶ The extensive knowledge of colloid behavior has formed a useful basis for understanding the fate of manufactured nanomaterials.

Natural colloids are subject to aggregation and sedimentation that results in particles in the size range 100–1000 nm.¹⁷ Aggregation is controlled by surface charge, particle size, ionic strength, pH and cation composition of the solution, and particle shape, which can affect steric interactions. Particles with near-neutral charge aggregate rapidly.

Attempts to use DLVO and other theoretical approaches developed for colloidal science to predict the aggregation of nanomaterials have been largely unsuccessful because of their polydispersity, shapes that frequently differ from spherical, and properties that vary from hydrophobic surfaces

with carbon-based materials, to core–shell structures of quantum dots, to magnetic particles in zerovalent iron, to metals and metal oxide materials.¹⁸

Any possible predictions are further complicated where surface coatings (surfactants, polymers, and polyelectrolytes) in some nanomaterial formulations are used to enhance dispersion stability. Depending on the nature of the surface binding, there is the potential for desorption in some media. Again, the important question will be how this stability is affected by an excess of natural colloidal particles. Some surface coatings may be susceptible to biodegradation, leading to nanoparticle aggregation.¹⁹

The expected environmental concentrations of manufactured nanomaterials in natural waters are typically below 20 µg/L (Table 1). Natural colloid concentrations are typically several orders of magnitude higher, ranging from 1 to 20 mg/L in freshwaters, higher in soil solutions, and marginally lower in seawater. Heteroaggregation with natural colloids is therefore likely to control the fate of most nanoparticles, yet despite this, most studies have focused on investigations of homoaggregation. Heteroaggregation has typically been considered for mixtures containing high concentrations of nanomaterials in the presence of low concentrations of natural colloids,^{20,21} largely driven by the relatively high detection limits of most of the techniques used to investigate

TABLE 1. Estimated Environmental Concentrations of Manufactured Nanomaterials Compared to Predicted No Effects Concentrations

nanomaterial	PEC, $\mu\text{g/L}$	water body	ref	PNEC, $\mu\text{g/L}$	ref
TiO ₂	0.7–16	freshwater	41	40	61
	<5–15	WWTP effluent	51	50	in 46
	1–10	freshwater	45		
	<8	Swiss rivers	47		
n-C ₆₀	0.0005–19	WWTP effluent	62	7.9	61
CNT	0.0005–0.0008	freshwater	41		
Ag	0.03–0.08	freshwater	41	40	41
	0.04–0.32	freshwater	45	1	in 46
	<0.03	Swiss rivers	47		
CeO ₂	0.1–1	freshwater	45	3000	in 46

particle size and/or aggregation kinetics, including dynamic light scattering, differential centrifugal separation, nanoparticle tracking analysis, and field flow fractionation.²²

To understand interactions with natural organic matter (NOM), numerous laboratory studies have investigated the effects of humic acid (HA) on manufactured nanomaterials, but few with the more relevant fulvic acid. The addition of standard Suwannee River HA greatly enhanced the dispersion of multiwalled carbon nanotubes in ultrapure deionized water,²³ with the same effects also seen in suspensions in Suwannee River water samples. The dispersion was greater than that observed in the presence of the surfactant sodium dodecylsulfate. The mechanism of the enhanced dispersion is likely to again involve both steric and electrostatic components, as was seen for natural colloids. Fullerene nanoparticle stability was also enhanced by HA, except in the presence of elevated ($>10^{-2}$ M) calcium chloride concentrations when calcium complexation resulted in intermolecular bridges involving HAs and fullerenes that enhanced aggregation.²⁴

Stabilization of iron oxide, alumina, titanium dioxide, gold, and Ag nanoparticles by HA has also been demonstrated,^{25–30} again, largely dependent on surface charge. The point of zero charge for iron oxide occurs near pH 7.8, with positively charged particles at low pH neutralized by HA. At higher pH, where the nanoparticles are negatively charged, adsorption of HA can still occur through hydrophobic interaction, with the result being an enhanced negative charge and enhanced stabilization, supplemented by a degree of steric stabilization. A unique finding was the role of HA in facilitating disaggregation of iron oxide particles at near neutral pH values.²⁵

Similar findings were obtained in studies of fulvic acid interactions with titanium dioxide nanoparticles.³¹ From experiments at mg/L concentrations of both, it was suggested that these nanoparticles might persist in natural waters, for longer than predicted by laboratory studies.

In natural systems, the ratio of NOM to nanomaterial concentration is likely to be higher than 10:1, with the interactions

less likely to be based on adsorption of NOM to the nanomaterials, but rather adsorption of NOM- or humic-coated nanomaterials to HA or NOM colloids, clusters, or aggregates. This phenomenon was demonstrated by Bae et al.³² for Ag nanomaterials of differing surface areas. The implication is that, despite the theoretical and laboratory-based predictions that humic acids (and by implication NOM) will generally lead to stabilization of environmental nanoparticles in aquatic systems (and in some instances involve disaggregation of aggregated nanoparticles), in the presence of environmental concentrations of natural colloids, that typically involve heterogeneous mixtures of NOM and inorganic binding phases, further adsorption of nanoparticles to these is likely, leading to the formation of larger and potentially unstable clusters.

The above discussion applies principally to freshwater systems of low ionic strength. In marine waters, most nanomaterials will be aggregated, unless sterically stabilized with surface coatings. This has been well demonstrated for citrate and polyvinylpyrrolidone (PVP) coated Ag nanoparticles.³³ Both particle types were negatively charged and stable over a wide range of salt concentrations. Consistent with DLVO theory, with increasing salinity (ionic strength), the electrophoretic mobility of the particles becomes less negative due to an increase in charge screening by the NaCl, to a point where the screening is complete and aggregation rates reach a maximum. The noncharged PVP coating assists in steric repulsion of particles, conferring greater stability against aggregation. This meant that slightly higher salt concentrations were required to induce aggregation, but this occurred below 0.1 M NaCl, about one fifth the salinity of seawater, compared to 0.03 M NaCl for the citrate-coated particles. The same study demonstrated the effect of humic acid in adding stability to the citrate-coated particles.

An excellent recent study by Quik et al.³⁴ demonstrated the significant role of heteroaggregation with natural colloids in the aggregation of nanomaterials (tested with CeO₂) in river waters, compared to greater stabilization by homoaggregation in filtered waters. Research attention is now being focused on the toxicity of nanomaterials in sediments, although sediment toxicity is likely to be less of an issue than aquatic toxicity.

The majority of the surface area and electrostatic charge in soils resides in the <1 μm size fraction.³⁵ Since the major part of the soil surface area is in the colloidal fraction, almost all surface-controlled processes, including adsorption reactions, nucleation, and precipitation/dissolution, involve colloids.³⁶ The importance of natural nanoparticles and colloids regarding the fate of manufactured nanoparticles

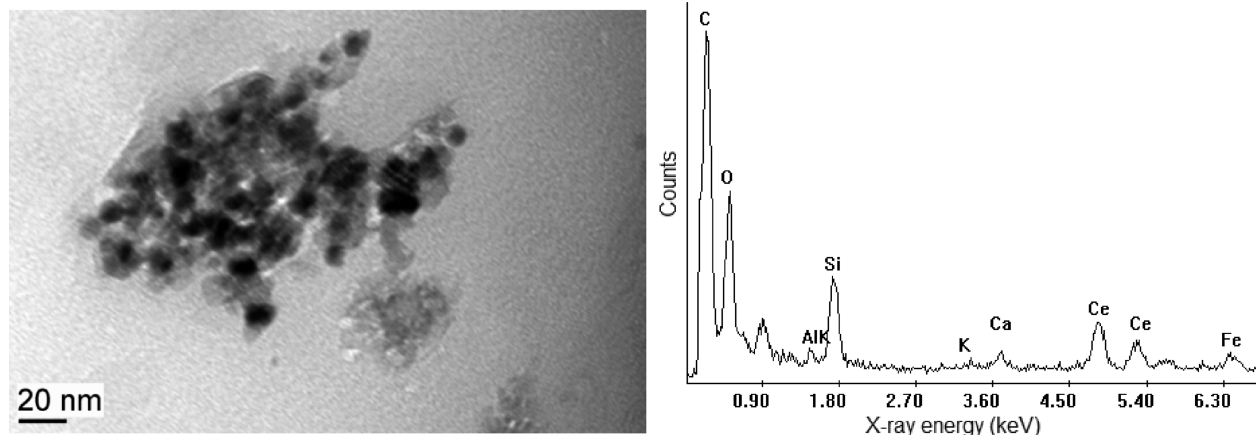


FIGURE 2. TEM image and energy dispersive (EDS) X-ray analysis of CeO₂ NPs aggregated with natural colloids in 0.45 μm filtrates of soil spiked with 12.6 mmol/kg CeO₂ NPs. The EDS spectra of the aggregates show peaks for Al, Ca, Fe, K, and Si that suggest that the aggregates also contain natural colloids (from Cornelis et al.²⁰).

in soils has been recognized.^{20,37,38} Cornelis et al.²⁰ investigated the retention (K_r) of CeO₂ nanoparticles (nominal particle size 20 nm) in soils and found a positive correlation with the clay content of soils and not with parameters that increase the homocoagulation rate of CeO₂ nanoparticles, suggesting that negatively charged CeO₂ nanoparticles were adsorbed preferentially by clay surfaces at positively charged sites. Examination of filtered soil suspensions using transmission electron microscopy (TEM) confirmed that heteroaggregation likely occurs (Figure 2).

Risk Assessment of the Environmental Impacts of Manufactured Nanomaterials

Given the limited availability of information on nanomaterial fate in the environment, the most appropriate assessment of risk is a simple comparison of predicted environmental concentrations (PECs) with predicted no effects concentrations (PNECs). The evaluation of both parameters presents major challenges.

Predicted Environmental Concentrations. The derivation of PECs requires the measurement or prediction of nanomaterial concentration and size, based on the fate and transport pathways once the nanomaterials are released to the environment. The possible pathways that will introduce nanomaterials to the environment are illustrated in Figure 3. With virtually no available measured environmental data, attempts have been made to model exposure.

The earliest life cycle assessment attempts suggested that direct entry of nanomaterials into aquatic systems via spillage, discharge, atmospheric deposition, or soil runoff was small compared to the fraction that proceeds to sewage treatment and might be in STP discharge waters.^{39–41}

In deducing mass flows, estimates of total product usage and release rates must be obtained, that can then be related to the time of exposure. Knowledge of the behavior of the nanomaterials in the environment (colloidal forms, attachment to particles, etc.) is used in coupled fate models to predict sediment/water partitioning during treatment and in the aquatic environment.

Boxall et al.⁴⁰ used a series of simple algorithms to predict the likely environmental concentrations from a limited range of manufactured nanomaterials in soils and waters. For waters, routes of entry included the following: direct entry into water bodies; inputs from spray drift of agrochemicals; runoff from contaminated soils; aerial deposition; and emissions from STPs. For soils, routes comprised the following: the application of remediation technologies; the application of plant protection products; the excretion of nanomedicines in veterinary products; aerial deposition; and the application of sewage sludge as a fertilizer.

The difficulty with some of these approaches is in covering the full range of potential sources of nanomaterials and not a limited selection, as well as reasonably estimating market usage.

Existing models of exposure for soluble contaminants have little applicability to nanoparticles, while the applicability to real systems of preliminary approaches to predictive modeling of the suspension stability and kinetics of aggregation of nanoparticles is, as yet, untested.⁴² Arvidsson et al.⁴³ acknowledged that the biggest limitation of exposure modeling remains a lack of understanding of the interactions of nanomaterials with NOM, and, where these have been studied, it has usually been under conditions that are not representative of the natural environment; however,

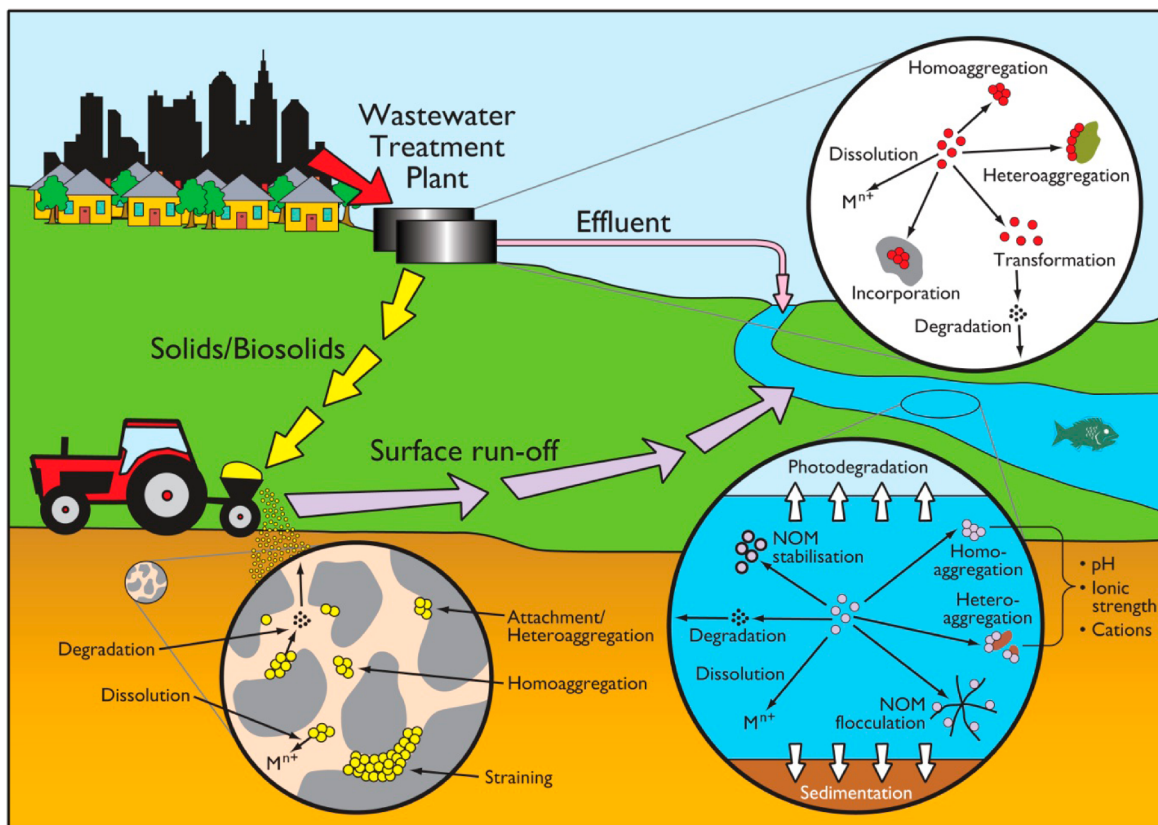


FIGURE 3. Pathways and transformations of nanomaterials in the environment.

other transformations, such as the sulfidation of silver, may be much more important

O'Brien and Cummins proposed a 3-level model to determine exposure risk,⁴⁴ beginning with laying out exposure-related concerns (level 1); determining particle characteristics (size, surface area, surface change, solubility, etc), behavior (aggregation, adsorption), and treatment (level 2); and relating these to exposure scenarios (level 3). This is a sound start, but trying to populate these levels with real environmentally relevant data again proved difficult.^{45,46} The approach ended up being highly subjective with difficulty in separating uncertainty from variability. The authors then defaulted to what amounted to a best professional judgment in a "black box" semiquantitative approach using "best available data" to rank exposure risk in aquatic systems. This was applied to nano-TiO₂ in paints, nanosilver in food packing, and nano-CeO₂ in fuels. Calculated exposure concentration ranges of 1–10 μg of TiO₂/L, 10–100 ng of Ag/L, and 0.1–1 μg of CeO₂/L were not dissimilar from the equally subjective predictions of other studies (Table 1).^{39,41,47}

The most recent estimates of nano TiO₂, Ag, and ZnO release, in Swiss rivers,⁴⁷ gave more detailed considerations to population density and river flow, in both a conservative

scenario with no agglomeration and an optimistic scenario with rapid and complete deposition. For TiO₂, concentrations typically ranged from 11 to 1620 ng/L for the conservative scenario and from 2 to 1620 ng/L for the optimum scenario. The corresponding estimates for ZnO and Ag were smaller again by factors of 14 and 240, respectively. There is an urgent need for data to validate the model predictions.

Coming out of the fate studies is the realization that, apart from accidental spillage of nanomaterials directly into waters or on land with subsequent washing into waters, wastewater treatment remains the largest source of nanomaterials either in the effluents or in the sludge which might be applied to land. Atmospheric deposition of nanomaterials is likely to be a small contributor to waters and soils, while, in developed countries at least, any effluents containing nanomaterials will require discharge licenses and will be more likely to go to a wastewater treatment plant.

Fate in Wastewater Treatment. Several studies have considered how nanomaterials partition during wastewater treatment. In the absence of suitable data, Mueller and Nowack assumed that 97% of the particles were removed, with 90% removal in a high exposure scenario,⁴¹ based largely on published data for the efficiency of removal of

particles $<0.1 \mu\text{m}$. It has been suggested that the presence of household or industrial detergents would result in the disaggregation of nanoparticles.⁴⁸ In a study of CeO_2 nanoparticles in a model wastewater treatment system, Limbach et al.⁴⁹ found that a small but significant fraction (6%) avoided aggregation and was released in the effluent (at 2–5 mg/L concentrations), largely as a result of stabilization in the presence of protein breakdown products and surfactants in the wastewater changing the ζ potential.

An examination of 10 wastewater treatment plants by Westerhoff et al.⁵⁰ showed removal of over 96% of titanium in influent sewage, with effluents typically containing $<25 \mu\text{g/L}$. Using TEM and energy dispersive X-ray analysis, spherical TiO_2 nanoparticles (crystalline and amorphous) of the order of 4 to 30 nm in diameter were detected in the effluents. An earlier study at one plant confirmed that tertiary filtration removed on average $82 \pm 21\%$ of TiO_2 as biosolids, leaving 10–50 $\mu\text{g/L}$ TiO_2 in the effluent.⁵¹ Nanoparticles were observed in the effluent but typically aggregated to several hundred nanometers in size.

Benn and Westerhoff noted that socks containing nanoparticulate Ag added as a deodorant released most of the Ag after repeated washings, with 70–90% in an ionic form, and the remainder as large nanoparticles (100–200 nm).⁵² In a simulated water-treatment process, all of the Ag was shown to be removable in the sludge, raising concerns about the impacts of application of sludge to land.

More recent studies of silver nanoparticles have demonstrated the presence of nanosized silver sulfide (Ag_2S) in sewage sludge products, formed by sulfidation reactions in sulfur-rich environments.⁵³ Both aggregated and nanosized Ag_2S were identified by high resolution TEM. Levard et al.⁵⁴ suggested that interaction of PVP-coated Ag nanoparticles with sodium sulfide resulted in strongly aggregated chain-like structures with Ag sulfide bridges between nanoparticles. It was proposed that the PVP desorbs and then readsorbs on the aggregated products.

The fate of hydrophobic nanoparticles and their aggregates (e.g., CNTs, fullerenes, quantum dots) may be modified through wastewater treatment if they undergo surface modifications to make them dispersible in water (e.g., carboxyl-conjugated quantum dots)⁵⁵ or are stabilized through HA adsorption.⁵⁶ There has been limited research into the fate and behavior of CNTs and quantum dots through wastewater treatment. Colloidal aggregates of fullerenes were effectively (up to 97%) removed in laboratory “jar tests”, by a series of alum coagulation, flocculation, sedimentation, and filtration processes, with an efficiency dependent

on the pH, alkalinity, organic matter content, and coagulant dosage.⁵⁷

The presence of nanoparticles in sewage sludge has implications for its use on land as a fertilizer, and very little work has been reported to date in this area. Biosolids contain both clay minerals and solid organic matter, that could bind nanoparticles strongly through heteroaggregation, and soluble organic matter, which could potentially mobilize nanoparticles through changes in surface charge. Cornelis et al.²⁰ found little difference in retention of CeO_2 nanoparticles to soils due to biosolids addition.

Several studies have indicated a potential for nanomaterials to interact with bacteria in sewage treatment plants. Choi et al.⁵⁸ showed that Ag nanoparticles were toxic to nitrifying bacteria and that this could imply detrimental effects to the microorganisms in wastewater treatment, with dissolved Ag responsible for the majority of the observed AgNP toxicity.^{58,59} Titanium dioxide nanoparticles in the presence of ultraviolet light were toxic to *E. coli* inhibiting the fouling of water treatment membranes.⁶⁰

Predicted No Effect Concentrations. Only limited toxicity data have been published that enable calculation of no effects concentrations and the development of reliable guidelines for nanomaterials in the environment.^{4,61} Most data exist for n- C_{60} and nano TiO_2 in aquatic systems, and it was possible to derive a chronic PNEC value of 7.9 $\mu\text{g/L}$ for n- C_{60} and 40 $\mu\text{g/L}$ for TiO_2 dispersed by sonication.⁶¹

A comparison of some of the estimated PEC values with the latest toxicity data for aquatic systems is given in Table 1. Again it is apparent that the environmental concentrations are well below levels of environmental concern. As yet, there are too few terrestrial toxicity studies to derive good threshold effects concentrations.

Ecological Risk Assessment. Despite the above uncertainties, a comparison of estimated exposure data with known toxicity data indicates that the PECs are currently orders of magnitude below those known to have environmental effects on aquatic biota (Table 1). In most countries, nanoparticle releases will ultimately be captured by wastewater treatment plants where they will partition to sludges, with a smaller concentration being released in the effluent and subject to dilution where these discharges enter rivers or coastal waters. Nanomaterials that escape treatment are likely to ultimately accumulate in benthic sediments, and their accumulation has yet to be fully investigated.

The application of biosolids or sludges to land is highly regulated in most countries through existing environmental guidelines.⁶³ Assessment of the potential ecological risks

from nanoparticles in land-applied biosolids may need a more detailed consideration of the life cycle and transformations of the nanomaterials during wastewater treatment. For example, the recently highlighted formation of highly insoluble Ag₂S in sewage sludge from Ag nanoparticles during wastewater treatment was shown to be an unexpected yet important immobilization pathway.⁵³

Where nanomaterials are stabilized by surface coatings, indications are that where these enter natural waters directly, they might nevertheless interact with larger colloidal suspensions.

Conclusions and Outlook

The overall findings of minimal environmental risk from manufactured nanomaterials should give regulatory agencies some reassurance, especially since the assumptions in estimations are conservative. This could change if nanomaterials find widespread use as fuel additives, pesticides, or other applications that lead to diffuse inputs. On-going research is therefore required to better define no-effect concentrations through increased toxicity testing. The challenge will be to undertake these and other fate studies in natural water, sediment, and soil systems at nanomaterial concentrations that are realistic and environmentally relevant, thus avoiding artifacts associated with very high concentrations.

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BIOGRAPHICAL INFORMATION

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FOOTNOTES

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REFERENCES

- Franklin, N. M.; Rogers, N. T.; Apte, S. C.; Batley, G. E.; Casey, P. E. Comparative toxicity of nanoparticulate ZnO, bulk ZnO and ZnCl₂ to a freshwater microalga (*Pseudokirchneriella subcapitata*): the importance of particle solubility. *Environ. Sci. Technol.* **2007**, *41*, 8484–8490.
- Wang, Y. G.; Li, Y. S.; Pennell, K. D. Influence of electrolyte species and concentration on the aggregation and transport of fullerene nanoparticles in quartz sands. *Environ. Toxicol. Chem.* **2008**, *27*, 1860–1867.
- French, R. A.; Jacobson, A. R.; Kim, B.; Isley, S. L.; Penn, R. L.; Baveye, P. C. Influence of ionic strength, pH, and cation valence on aggregation kinetics of titanium dioxide nanoparticles. *Environ. Sci. Technol.* **2009**, *43*, 1354–1359.
- Klaine, S. J.; Alvarez, P. J. J.; Batley, G. E.; Fernandes, T. F.; Handy, R. D.; Lyon, D.; Mahendra, S.; McLaughlin, M. J.; Lead, J. R. Nanomaterials in the environment: behaviour, fate, bioavailability and effects. *Environ. Toxicol. Chem.* **2008**, *27*, 1825–1851.
- ISO. Nanotechnologies—Terminology and definitions for nano-objects—Nanoparticle, nanofibre and nanoplate, ISO/TS 27687, 2008 (E). International Organization for Standardization: Geneva, Switzerland, 4 pages.
- Abraham, M. H.; Green, C. E.; Acree, W. E. Correlation and prediction of the solubility of buckminsterfullerene in organic solvents; estimation of some physicochemical properties. *J. Chem. Soc., Perkin Trans.* **2000**, *2*, 281–286.
- Sayes, C. M.; Fortner, J. D.; Guo, W.; Lyon, D.; Boyd, A. M.; Ausman, K. D.; Tao, Y. J.; Sitharaman, B.; Wilson, L. J. The differential cytotoxicity of water-soluble fullerenes. *Nano Lett.* **2004**, *4*, 1881–1887.
- Rogers, N. J.; Franklin, N. M.; Apte, S. C.; Spadaro, D.; Angel, B.; Batley, G. E.; Lead, J. R.; Baalousha, M. Behaviour and toxicity to algae of nanoparticulate CeO₂ in freshwater. *Environ. Chem.* **2010**, *7*, 50–60.
- Derfus, A. M.; Chan, W. C. W.; Bhatia, S. N. Probing the nanotoxicity of semiconductor quantum dots. *Nano Lett.* **2004**, *4*, 11–18.
- ANZECC/ARMCANZ. Australian and New Zealand guidelines for fresh and marine water quality. Australian and New Zealand Environment and Conservation Council/Agriculture and Resource Management Council of Australia and New Zealand, Canberra, ACT, Australia, 2000.
- Liu, J. Y.; Hurt, R. H. Ion release kinetics and particle persistence in aqueous nano-silver colloids. *Environ. Sci. Technol.* **2010**, *44*, 2169–2175.
- Canesi, L.; Ciacci, C.; Fabbri, R.; Marcomini, A.; Pojana, G.; Gallo, G. Bivalve molluscs as a unique target group for nanoparticle toxicity. *Mar. Environ. Res.* **2012**, *76*, 16–21.
- Holbrook, D. R.; Murphy, K. E.; Morrow, J. B.; Cole, K. D. Trophic transfer of nanoparticles in a simplified invertebrate food web. *Nat. Nanotechnol.* **2008**, *3*, 352–355.
- Zhu, X.; Wang, J.; Zhang, X.; Chang, Y.; Chen, Y. Trophic transfer of TiO₂ nanoparticles from daphnia to zebrafish in a simplified freshwater food chain. *Chemosphere* **2010**, *79*, 928–933.
- Judy, J. D.; Unrine, J. M.; Bertsch, P. M. Evidence for biomagnification of gold nanoparticles within a terrestrial food chain. *Environ. Sci. Technol.* **2011**, *45*, 776–781.
- Wilkinson, K. J.; Lead, J. R. *Environmental Colloids and Particles: Behaviour, Separation and Characterisation*; John Wiley and Sons: Chichester, U.K., 2007.
- Buffle, J.; Leppard, G. G. Characterisation of aquatic colloids and macromolecules. 2. Key role of physical structures on analytical results. *Environ. Sci. Technol.* **1995**, *29*, 2176–2184.
- Hotze, E. M.; Phenrat, T.; Lowry, G. V. Nanoparticle aggregation: Challenges to understanding transport and reactivity in the environment. *J. Environ. Qual.* **2010**, *39*, 1909–1924.
- Kirschling, T. L.; Golas, P. L.; Unrine, J. M.; Matyjaszewski, K.; Gregory, K. B.; Lowry, G. V.; Tilton, R. D. Microbial bioavailability of covalently bound polymer coatings on model engineered nanomaterials. *Environ. Sci. Technol.* **2011**, *45*, 5253–5259.
- Cornelis, G.; Ryan, B.; McLaughlin, M. J.; Kirby, J. K.; Beak, D.; Chittleborough, D. Solubility and batch retention of CeO₂ nanoparticles in soils. *Environ. Sci. Technol.* **2011**, *45*, 2777–2782.

- 21 Yates, P. D.; Franks, G. V.; Jameson, G. J. Orthokinetic heteroaggregation with nanoparticles: Effect of particle size ratio on aggregate properties. *Colloids Surf., A* **2008**, *326*, 83–91.
- 22 Baalousha, M.; Stolpe, B.; Lead, J. R. Flow field-flow fractionation for the analysis and characterization of natural colloids and manufactured nanoparticles in environmental systems: A critical review. *J. Chromatogr., A* **2011**, *1218*, 4078–4103.
- 23 Hyung, H.; Fortner, J. D.; Hughes, J. B.; Kim, J.-H. Natural organic matter stabilization of multi-walled nanotubes in water. *Environ. Sci. Technol.* **2007**, *41*, 179–184.
- 24 Chen, K. L.; Elimelech, M. Influence of humic acid on the aggregation kinetics of fullerene (C60) nanoparticles in monovalent and divalent electrolyte solutions. *J. Colloid Interface Sci.* **2007**, *309*, 126–34.
- 25 Baalousha, M. Aggregation and disaggregation of iron oxide nanoparticles: Influence of particle concentration, pH and natural organic matter. *Sci. Total Environ.* **2009**, *407*, 2093–2101.
- 26 Saleh, N.; Kim, H. J.; Phenrat, T.; Matyjaszewski, K.; Tilton, R. D.; Lowry, G. V. Ionic strength and composition affect the mobility of surface-modified Fe⁰ nanoparticles in water-saturated sand columns. *Environ. Sci. Technol.* **2008**, *42*, 3349–3355.
- 27 Tipping, E.; Higgins, D. C. The effect of adsorbed humic substances on the colloid stability of haematite particles. *Colloids Surf.* **1982**, *5*, 85–92.
- 28 Baalousha, M.; Manciuola, A.; Cumberland, S.; Kendall, K.; Lead, J. R. Aggregation and surface properties of iron oxide nanoparticles: influence of pH and natural organic matter. *Environ. Toxicol. Chem.* **2008**, *27*, 1875–1882.
- 29 Yang, K.; Lin, D.; Xing, B. Interactions of humic acid with nanosized inorganic oxides. *Langmuir* **2009**, *25*, 3571–3576.
- 30 Diegoli, S.; Manciuola, A. L.; Begum, S.; Jones, I. P.; Lead, J. R.; Preece, J. A. Interaction between manufactured gold nanoparticles and naturally occurring organic macromolecules. *Sci. Total Environ.* **2008**, *402*, 51–61.
- 31 Domingos, R.; Baalousha, M. A.; Yon, J.-N.; Lead, J. R.; Leppard, G. G.; Wilkinson, K. J. Characterizing manufactured nanoparticles in the environment: multimethod determination of particle sizes. *Environ. Sci. Technol.* **2009**, *43*, 7277–7284.
- 32 Bae, E.; Park, H.-J.; Yoon, J.; Kim, Y.; Choi, K.; Yi, J. Bacterial uptake of silver nanoparticles in the presence of humic acid and AgNO₃. *Korean J. Chem. Eng.* **2011**, *28*, 267–271.
- 33 Huynh, K. A.; Chen, K. L. Aggregation kinetics of citrate and polyvinylpyrrolidone coated silver nanoparticles in monovalent and divalent electrolyte solutions. *Environ. Sci. Technol.* **2011**, *45*, 5564–5571.
- 34 Quik, J. T. K.; Stuart, M. C.; Wouterse, M.; Peijnenburg, W.; Hendriks, A. J.; Van de Meent, D. Natural colloids are the dominant factor in the sedimentation of nanoparticles. *Environ. Toxicol. Chem.* **2012**, *31*, 1019–1022.
- 35 Borkovec, M.; Wu, Q.; Degovics, G.; Laggner, P.; Sticher, H. Surface area and size distributions of soil particles. *Colloids Surf., A* **1993**, *73*, 65–76.
- 36 Goldberg, S. Soil Colloidal Behaviour. In *Handbook of Soil Science*; Sumner, M. E., Ed.; CRC Press: Boca Raton, London, New York, Washington, DC, 2000; Chapter 6, pp B-195 to B-240.
- 37 Fang, J.; Shan, X.; Wen, B.; Lin, J.; Owens, G. Stability of titania nanoparticles in soil suspensions and transport in saturated homogenous soil columns. *Environ. Pollut.* **2009**, *157*, 1101–1109.
- 38 Cornelis, G.; Kirby, J. K.; Beak, D.; Chittleborough, D.; McLaughlin, M. A method for determination of retention of silver and cerium oxide manufactured nanoparticles in soils. *Environ. Chem.* **2010**, *7*, 298–308.
- 39 Blaser, S. A.; Scheringer, M.; MacLeod, M.; Hungerbühler, K. Estimation of cumulative aquatic exposure and risk due to silver: Contribution of nano-functionalized plastics and textiles. *Sci. Total Environ.* **2008**, *390*, 396–409.
- 40 Boxall, A. B. A.; Chaudhry, Q.; Sinclair, C.; Jones, A.; Aitken, R.; Jefferson, B.; Watt, C. Current and future predicted environmental exposure to engineered nanoparticles. Central Science Laboratory Report, University of York, prepared for the UK Department of Environment, Food and Rural Affairs, 2007.
- 41 Mueller, N. C.; Nowack, B. Exposure modeling of engineered nanoparticles in the environment. *Environ. Sci. Technol.* **2008**, *42*, 4447–4453.
- 42 Mackay, C. E.; Johns, M.; Salatas, J. H.; Bessinger, B.; Perri, M. Stochastic probability modelling to predict the environmental stability of nanoparticles in aqueous suspension. *Integr. Environ. Assess. Manage.* **2006**, *2*, 293–298.
- 43 Arvidsson, R.; Molander, S.; Sanden, B. A.; Hasselöv, M. Challenges in exposure modelling of nanoparticles in aquatic environments. *Hum. Ecol. Risk Assess.* **2011**, *17*, 245–262.
- 44 O'Brien, N. J.; Cummins, E. J. Development of a three-level risk assessment strategy for nanomaterials. In *Nanomaterials, Risks and Benefits*; Linkov, I., Steevens, J., Eds.; NATO Science for Peace and Security Series C, Environmental Security; Springer: London, 2009; pp 161–178.
- 45 O'Brien, N. J.; Cummins, E. J. Ranking initial environmental and human health risk resulting from environmentally relevant nanomaterials. *J. Environ. Sci. Health, A* **2010**, *45*, 992–1007.
- 46 O'Brien, N. J.; Cummins, E. J. A risk assessment framework for assessing metallic nanomaterials of environmental concern: Aquatic exposure and behavior. *Risk Anal.* **2011**, *31*, 706–726.
- 47 Gottschalk, F.; Ort, C.; Scholz, R. W.; Nowack, B. Engineered nanomaterials in rivers—Exposure scenarios for Switzerland at high spatial and temporal resolution. *Environ. Pollut.* **2011**, *159*, 3439–3445.
- 48 Fernandes, M.; Rosenkranz, P.; Ford, A.; Christofi, N.; Stone, V. Ecotoxicology of nanoparticles (NPs). *SETAC Globe* **2006**, 43–45.
- 49 Limbach, L. K.; Bereiter, R.; Müller, E.; Krebs, R.; Gälli, R.; Stark, W. J. Removal of oxide nanoparticles in a model wastewater treatment plant: influence of agglomeration and surfactants on clearing efficiency. *Environ. Sci. Technol.* **2008**, *42*, 5828–5833.
- 50 Westerhoff, P.; Song, G.; Hristovski, K.; Kiser, M. A. Occurrence and removal of titanium at full scale wastewater treatment plants: implications for TiO₂ nanomaterials. *J. Environ. Monit.* **2011**, *13*, 1195–1203.
- 51 Kiser, M. A.; Westerhoff, P.; Benn, T.; Wang, Y.; Perez-Rivera, J.; Hristovski, K. Titanium nanomaterial removal and release from wastewater treatment plants. *Environ. Sci. Technol.* **2009**, *43*, 6757–6763.
- 52 Benn, T. M.; Westerhoff, P. Nanoparticle silver released into water from commercially available sock fabrics. *Environ. Sci. Technol.* **2008**, *42*, 4133–4139.
- 53 Kim, B.; Park, C.-S.; Murayama, M.; Hochella, M. F. Discovery and characterization of silver sulfide nanoparticles in final sewage sludge products. *Environ. Sci. Technol.* **2008**, *44*, 7509–7514.
- 54 Levard, C.; Reinsch, B. C.; Michel, F. M.; Oumahi, C.; Lowry, G. V.; Brown, G. E. Sulfidation processes of PVP-coated silver nanoparticles in aqueous solution: impact on dissolution rate. *Environ. Sci. Technol.* **2011**, *45*, 5260–5266.
- 55 Al-Salim, N.; Barraclough, E.; Burgess, E.; Clothier, B.; Deurer, M.; Green, S.; Malone, L.; Weir, G. Quantum dot transport in soil, plants, and insects. *Sci. Total Environ.* **2011**, *409*, 3237–3248.
- 56 Hyung, H.; Fortner, J. D.; Hughes, J. B.; Kim, J. H. Natural organic matter stabilizes carbon nanotubes in the aqueous phase. *Environ. Sci. Technol.* **2007**, *41*, 179–184.
- 57 Hyung, H.; Kim, J. Dispersion of C₆₀ in natural water and removal by conventional drinking water treatment processes. *Water Res.* **2009**, *43*, 2463–2470.
- 58 Choi, O.; Deng, K. K.; Kim, N.-J.; Ross, L.; Surampalli, R. Y.; Hu, Z. The inhibitory effects of silver nanoparticles, silver ions, and silver chloride colloids on microbial growth. *Water Res.* **2008**, *42*, 3066–3074.
- 59 Radniecki, T.; Stankov, D. P.; Neigh, A.; Nason, J. A.; Semprini, L. Influence of liberated silver from silver nanoparticles on nitrification inhibition of *Nitrosomonas europaea*. *Chemosphere* **2011**, *85*, 43–49.
- 60 Kwak, S. Y.; Kim, S. H.; Kim, S. S. Hybrid organic/inorganic reverse osmosis (RO) membrane for bactericidal anti-fouling. 1. Preparation and characterization of TiO₂ nanoparticle self-assembled aromatic polyamide thin-film-composite (TFC) membrane. *Environ. Sci. Technol.* **2001**, *35*, 2388–2394.
- 61 Batley, G. E.; McLaughlin, M. J. Fate of manufactured nanomaterials in the Australian environment. CSIRO Niche Manufacturing Flagship Report, 2008; Lucas Heights, NSW, Australia, 85 pages.
- 62 Farre, M.; Perez, K.; Gajda-Schranz, K.; Osorio, V.; Kantiani, L.; Ginebreda, A.; Barcelo, D. First determination of C60 and C70 fullerenes and N-methylfulleropyrrolidine C60 on the suspended material of wastewater effluents by liquid chromatography hybrid quadrupole linear ion trap tandem mass spectrometry. *J. Hydrol.* **2010**, *383*, 44–51.
- 63 US EPA. United States Environmental Protection Agency, Federal Register Part II. 40 CFR Part 257. Standards for the Use or Disposal of Sewage Sludge: Final Rules; US EPA: Washington, DC, 1993; 348 pages.