

Book of Abstracts

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From nanoparticle surface chemistry to ecotoxicology

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Given the predictability for some nanoparticles (NP) to enter aquatic environments, associated potential risks will depend on their bioavailability and ecotoxicological hazard potential. The large heterogeneity of particles with regard to chemical composition, size, morphology and physicochemical characteristics on one hand and the large absence of experimental data on the other hand, clearly demand for a strategy to prioritize possible research questions. In spite of these complexities, basic principles from surface chemistry and ecotoxicology represent main elements to draw on for formulating predictive hypothesis that help on guiding environmental research on fate and effects of particles. The surface properties of NP are determinant of their aggregation behavior, chemical reactivity and thus of their interaction with chemical components of natural waters. Therefore, the bioavailability of nanoparticles will be modified according to the interactions with natural organic matter, hydrophobic organic species and metals. Biological interfaces constitute the primary site for interaction between bioavailable particles and organisms. Consideration of the main biological transport systems and of sieving properties of membranes allows for predictions on whether NP will be taken up and internalized in cells. Finally, advances in understanding biochemical interactions of chemical pollutants with aquatic organisms offer an array of endpoints for examining toxic effects of NP. Here we exemplify this process for the case of metal nanoparticles of different sizes and their interactions with selected aquatic organisms.

Life-cycle perspectives of nanoparticle-containing products

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Assessing the risks of engineered nanoparticles in the environment requires an understanding of their mobility, reactivity, bioavailability, persistency and toxicity. A lot of research is currently devoted to these topics. This basic knowledge is needed but is not sufficient to allow for a realistic risk assessment of nanoparticles. It is also necessary to make an evaluation of the expected quantities and concentrations of nanoparticles in environmental and technical systems. To date almost nothing is known about these issues, neither from an analytical point of view (e.g. actual measurements of nanoparticles) nor with respect to theoretical or modeling studies. We assessed potential forms and quantities of released nanoparticles (exposure scenarios) from a life-cycle perspective of nanoparticle-containing products and modeled the expected concentrations of nanoparticles in environmental and technical media. The focus was mainly on three types of engineered nanoparticles: nano-Ag, nano-TiO₂ and carbon nanotubes. In addition the emissions of engineered and unintentionally produced nanoparticles to soils were assessed. The quantification was based on a substance flow analysis from products to air, soil and water in Switzerland. The following parameters were used as model inputs: estimated production volume, allocation of the production volume to product categories, particle release from products and flow coefficients within the technical and environmental compartments. The predicted environmental concentrations (PEC) were then compared to the predicted no effect concentrations (PNEC), derived from literature, to estimate a possible risk. The expected concentrations of the nanoparticles in the different environmental compartments vary widely, caused by the different life cycles of the nanoparticle-containing products. The PEC values for nTiO₂ in water are 0.7 - 16 µg/l and close to or higher than the PNEC value for nTiO₂ (<1 µg/l). The risk quotients (PEC/PNEC) for CNT and nAg were much smaller than one and therefore no adverse effect are currently expected for those particles. The results of this study allow for the first time to carry out a quantitative risk assessment of nanoparticles in the environment and call for further detailed studies of nTiO₂.

Swiss action plan on synthetic nanomaterials

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In Spring 2006 work on the Swiss Action Plan was officially launched. A work package is developed in collaboration with experts and stakeholders in order to identify critical applications of manufactured nanomaterials and to minimize possible detrimental effects on human health and the environment. The coordination of this work package with international organizations (OECD, ISO) and EU is important. The promotion of safety research on manufactured Nanomaterials and the dialog with the public and stakeholders are other objectives of the Action Plan. The first deliverable of the action plan was a basic report, published in July 2007 (<http://www.umwelt-schweiz.ch/nanotechnologie>), containing an overview of the current knowledge about the risks of manufactured nanoparticles. Topics discussed are human toxicity and ecotoxicity of nanomaterials, occupational health and safety, regulation and standardisation, the assessment of the consequences of technology and communication. Finally, a list of risk research needs is presented.

Present Swiss legislation does not take the specific properties of manufactured nanomaterials into account. The action plan of Switzerland covers the following issues:

- Product information for consumers
- Guidance for self-supervision
- Information about manufactured nanomaterials on the material safety data sheets
- Maximum tolerable concentrations at the workplace
- Regulation of waste disposal
- Obligations to report, notify or authorisation (test requirements, special procedures for the registration of nanoparticles)
- Limitation of emissions into the environment
- Regulation on major accidents (tonnage thresholds)
- Development and market launch of sustainable applications of nanotechnology

Exposure Analysis of Engineered Nanoparticles in the Environment

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It is inevitable that during their manufacture and use, engineered nanoparticles (ENPs) will be released to the environment. While ENPs may be emitted during the manufacturing process, the route of input to the environment will primarily depend on the end use of the ENP-containing product. Available data indicate that, following release to water and air, nanoparticles will aggregate to some degree and that the behaviour of the resulting aggregates will be very different from the free nanoparticle. The degree of aggregation and the size range of the aggregates is dependent on the characteristics of the particle, the concentration of the particles and the characteristics of the environmental system. ENPs will exhibit differing mobilities in the soils and waterbodies and in water treatment processes compared to their corresponding parent form. The behaviour of nanoparticles in environmental systems is therefore highly complex and appears to be dependent on not only the particle type but also the particle size and the nature of the receiving environments. As a result of the lack of data on usage and environmental fate, a framework of simplistic models and algorithms for estimating concentrations in water, soil and air has been developed and applied to a range of ENPs. This modelling framework was applied to estimate the likely concentrations of ENPs in water and soil for a range of usage scenarios. Concentrations of silver, aluminium and fullerene C60 concentrations were predicted to be in the ng/l, whereas, titanium dioxide, zinc oxide, nanolatex and hydroxyapatite are predicted to be in the µg/l range. Predicted concentrations in soil ranged from <0.01 (cerium dioxide) to around 4.3 mg/kg (nanolatex). Predictions were also obtained for concentrations of selected ENPs in the air compartment. If, in the future, all of the product types investigated contained engineered nanoparticles, then concentrations in water could range from < 1 ng/l (cerium dioxide) to 1 mg/l (nanolatex). These exposure data provide a benchmark to a) inform the development of new analytical methodologies for environmental system, b) inform the design of environmental fate studies; and c) interpret the significance of existing ecotoxicology data on ENPs. A comparison of the results of the exposure estimations with the available ecotoxicological data is reassuring and indicates that even the conservative exposure concentrations generated in this study are many orders of magnitude lower than concentrations likely to cause acute effects in invertebrates, fish algae or sublethal effects on fish, invertebrates or bacteria. Whilst this study has identified the potential environmental exposure arising from a range of key ENP types, the assessment has been limited by the availability of data and knowledge. Work in the future should therefore focus on 1) establishing a detailed knowledge of the content and use of products containing ENPs in the UK; 2) developing an understanding of the factors and processes affecting the fate and transport of ENPs in the environment; 3) the development and evaluation of more complex exposure assessment models; and 4) the development of a better understanding of the ecotoxicity of ENPs under environmentally-relevant exposure situations.

Nanoinventory: a fact-based model of the occupational exposure to manufactured nanoparticles and the maximal release potential into the environment in Swiss industry

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A large number of applications using manufactured nanoparticles <100 nm are entering industrial processes and consumer products. Manufactured nanoparticles might cause similar negative health effects to micro- and nano-sized ambient particles, and many potential effects on ecosystems are still unassessed.

We plan to create a quantitative model for the occupational exposure and the maximal release potential of nanoparticles in the Swiss industry by using:

a) A telephone survey of two hundred companies helped us to test the feasibility of a survey in the Swiss industry, and the quality, which can be achieved. The study gave us valuable information about the knowledge of safety-managers concerning the handling of nanoparticles in their companies. Several types of nanoparticles were reported to be used in quantities of more than 1000 kg/annum per company, but the majority of nanoparticle applications were small production scale. Most of the companies had many open questions about best practices: this shows again that guidelines and protection strategies should be developed soon.

b) These results allowed us to formulate the currently ongoing, detailed, representative survey on a national level. It is a questionnaire-based estimation of the potentially exposed employees, containing also questions about the amount and type of used particles to explore the maximal release potential. It reflects the widest possible range of different Swiss industries. This questionnaire-based survey will be completed in a second survey-round with information about the real exposure by a workplace exposure assessment in selected companies and for selected application types. The assessment in the identified companies will produce better estimates of the quantity of nanoparticles used by each company and verify/estimate their permanent and maximal emission into environment.

c) These potential/representative and real/selective datasets will be used to build a model of the potential release of nanoparticles and the occupational exposure to them in Swiss industry. The chosen approach will be valuable for policy makers as well as for health, safety and environmental managers in industry: the information will be fact-based and statistically representative for the whole country. The data about occupational exposure and potential release can potentially be used as an important element for risk evaluations and prevention strategies.

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Fate and transport of ionic and nanoparticle silver released from commercially available socks

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The application of nanotechnology in many consumer products¹ makes it now possible to begin quantifying the exposure of our surroundings to engineered nanomaterials. Manufacturers of clothing articles (e.g., socks) employ nanosilver (n-Ag) as an antimicrobial agent to minimize the microbial growth that causes foot odor. However, the impacts to human health and various ecosystems from the release of n-Ag from commercial products are unknown²⁻⁴. A need exists for qualitative analyses of n-Ag released from consumer products to evaluate the risk of this new commercial market. Six types of socks, alleged to contain n-Ag, were characterized and washed with water to assess the potential release of silver (ionic and colloidal) into domestic wastewaters. Upon acid-digestion, the silver contents of the socks varied considerably, and were less than 1360 $\mu\text{g-Ag} / \text{g-sock}$. Microscopy conducted on ashed (550°C) sock material revealed the presence of silver nanoparticles ranging in diameter from 10 to 500 nanometers. After being washed in 500 mL of distilled water, some socks leached as much as 650 μg of silver. Filtration and ion selective electrode (ISE) methods suggest that silver is leached in colloidal and ionic forms. The ionic-Ag portion varied greatly between the wash waters of two sock types, which suggests that the release of n-Ag from socks as colloidal or ionic silver is controlled either by the n-Ag properties or the sock manufacturing process. Finally, the adsorption of the leached silver to wastewater treatment biomass was investigated. Based upon biosolids sorption tests of silver, a model for an activated sludge wastewater treatment plant (WWTP) suggests that a typical wastewater treatment facility could treat an influent concentration of 2900 ppb of leached silver before exceeding the 1.9 ppb EPA salt water quality criteria level. For this level of influent silver, the wasted activated sludge would contain over 30,000 ppb of silver, which could be reintroduced to the environment through agricultural land-application of WWTP biosolids.

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Natural and Anthropogenic Environmental Nanoparticulates: Their Microstructural Characterization and Respiratory Health Implications

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A wide range of environmental particulate matter (PM) both indoor and outdoor consisting of natural and anthropogenic PM collected by high volume air filters, electrostatic precipitation, and thermophoretic precipitation directly onto transmission electron microscope (TEM) coated grid platforms has been systematically characterized by TEM, energy dispersive X-ray spectrometry (EDS) and scanning electron microscopy (SEM) (including scanning transmission electron microscopy (STEM)). 93% of outdoor PM₁ (<1 μm aggregate diameter) are crystalline; >80% are aggregated. >60% of primary components are <100 nm diameter. 40% of PM₁ are carbonaceous (including multiwall carbon nanotubes (MWCNTs) and multiconcentric fullerenes) soot PM, >30% contain Si, >15% contain Fe (dominated by Fe₂O₃ and Fe₃O₄) and ~10% contain Ti (as TiO₂). Aggregates of MWCNTs and nanosilica (~6 nm) are common in the outdoor air and have also been observed in a 10,000 year-old Greenland ice core melt water sample.

Multiply-replicated cytotoxicity (*in vitro*) assays utilizing murine and human macrophage cell lines as well as a human epithelial (lung model) cell line (A549) consistently demonstrate varying degrees of cell death for essentially all nanoparticulate, aggregated PM, especially indoor and outdoor generated soot PM and black carbon (BC) (~30% of tire composition and tire wear debris world-wide): candle soot, wood soot, diesel soot, tire soot, etc.; with primary, spherule, nanoparticulate diameters ranging from 15 to 80 nm. Soot from natural gas combustion exhibits the most toxicity in the smallest mass concentrations along with BC. No correlation of cytotoxic response with PAH content of soot PM was observed and no correlation with specific surface area for a variety of nano-PM aggregate species has been observed. Cytokine release (respiratory-related IL-6 and IL-8) was detected for Fe₂O₃, chrysotile asbestos, BC, and MWCNT PM while reactive oxygen species (ROS) production has been detected for Fe₂O₃, asbestos, BC, and MWCNT aggregate PM as well as natural gas combustion PM (consisting of soot PM and MWCNT aggregate mixtures). Taken together, these results illustrate proinflammatory and related respiratory health issues for a wide range of natural and anthropogenic environmental nano-PM aggregates, but fundamental mechanisms associated with these issues are largely unknown.

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Reactivity of iron and iron oxide nanoparticles

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Elucidating the fundamental chemical, structural, and physical properties can shed new and unexpected light on the behavior of both natural and synthetic nanoparticles. Iron oxides and oxyhydroxides commonly occur as nanoparticles in the nanosize range and strongly impact the biogeochemical cycle of iron and other species at and near the Earth's surface. We use a range of techniques in order to elucidate the link between chemical reactivity and the physical and structural properties of iron oxide nanoparticles. For example, results from experiments quantifying the rates of reductive dissolution of ferrihydrite nanoparticles of varying size and composition but similar aspect ratio will be discussed. Overall, results demonstrate little to no size dependence in the relative reactivity, as quantified by rate constants that are normalized to specific surface area by way of the empirical rate law for reductive dissolution, of the ferrihydrite nanoparticles. Similarly, no size dependence in the activation energy for reductive dissolution was observed. In contrast, a marked increase in the pre-exponential factor of the Arrhenius equation as a function of decreasing particle size was observed, and this increase significantly outpaced what would be predicted by simple increase in specific surface area. This result is consistent with a hypothesis that the ordering of water molecules at the nanoparticle surface mediates diffusion to the surface and is size dependent. Furthermore, sample preparation and storage is known to strongly impact the properties of nanoparticles. Results comparing the reactivity of nanoparticles as a function of age and storage condition will be specifically addressed. In general, dried powders are substantially less reactive than are fresh, never-dried suspensions. Solid-state characterization methods include X-ray diffraction, X-ray absorption, high-resolution transmission electron microscopy, and low temperature magnetism.

Effects of Surface Oxidation on the Behavior of Carbon Nanotubes in Aquatic Environments

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The environmental implications of carbon nanotubes (CNTs) in the environment will be strongly dependent upon both surface and solution properties, which will control not only the particles' behavior with respect to aggregation and transport (sedimentation and deposition) but also their behavior with respect to the sorption of other potentially toxic dissolved species, such as anthropogenic organic chemicals and dissolved metal species. The combination of these properties will control, for example, whether CNTs serve to immobilize other toxic contaminants or facilitated their transport. In the latter regard, there is also some concern that the small diameter of CNTs may also allow them to interact with biological cells and membranes in unusual ways, perhaps allowing them to act as "Trojan horses" that can carry other contaminants into organisms. In the work described here, we seek to understand the effects of both aqueous conditions and CNT surface chemistry on CNT behavior in aquatic environments. This same information can also be used to tailor the design and manufacture of CNTs toward those with more desirable transport or sorption properties.

Oxygen is often grafted onto the surfaces of CNTs soon after their manufacture, either incidentally as a consequence of purification processes or deliberately in order to facilitate their subsequent handling and application. Surface functionalization can also occur during exposure to oxidizing agents after release to the environment. In recent work, we have oxidized the surfaces of a series of MWCNTs in a controlled manner with several different oxidants, including HNO₃, H₂O₂ and KMnO₄. For each treatment, the extent of oxidation was quantified using X-ray photoelectron spectroscopy (XPS) and for selected cases, specific oxygen-containing functional groups were quantified using XPS in conjunction with fluorinating agents that react selectively with specific surface groups (e.g. hydroxyl and carboxylic acid). Oxidized CNTs were then tested with respect to (1) their ability to adsorb aqueous solutes (low molecular weight toxic organic contaminants such as naphthalene and divalent metal cations such as Zn²⁺ and Cd²⁺); and (2) their stability in aqueous solution, as measured by monitoring aqueous suspensions over time with both UV-vis spectroscopy and dynamic light scattering. CNT transport studies through porous media are also planned.

Our results show how oxygen-containing functional groups on CNT surfaces will influence the CNT properties in water. More highly oxidized CNTs remain stable over a much wider range of aquatic conditions (defined by pH, ionic strength and concentration of Ca²⁺) than do pristine forms, following a well-defined functional relationship. Sorption characteristics are also strongly affected by surface oxidation. For naphthalene, the sorption capacity of pristine (as received) MWCNTs was intermediate between that of a natural char and an activated carbon, but surface oxidation strongly decreased the CNTs' sorption capacity, in again a well-defined way, with small (~10%) increases in surface oxygen causing much larger (~70%) decreases in maximum adsorption capacity. Highly nonlinear isotherms were observed, with linearity found to be relatively insensitive to surface oxidation, as characterized by Freundlich isotherm exponents. For divalent metal contaminants such as Zn²⁺, the opposite trend was observed, as expected on the basis of electrostatic interactions. For the divalent metals, Langmuir isotherm models adequately describe the data – both sorption affinity and capacity were found to be higher for more highly oxidized CNTs.

Effects of solution chemistry on C₆₀ aggregate formation and transport

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Despite the rapid growth in the nanotechnology field, little is currently known about the unintended health or environmental effects of manufactured nanomaterials. Because experience with naturally occurring nanoscale particles present in air has shown that they are hazardous to human health and that they can easily travel global-scale distances in the atmosphere, such scenarios involving engineered nanoparticles must be explored. This project seeks to examine carbonaceous nanomaterial fate and transport in the environment. In particular, we are interested in how these particles behave when transferred from water to air or vice versa.

The discovery that negatively charged aggregates of C₆₀ are stable in aqueous environments has elicited concerns regarding the potential environmental and health effects of these aggregates. Although many previous studies have used aggregates synthesized using intermediate organic solvents, this work employed an aggregate production method believed to more closely emulate the fate of fullerene upon accidental release – extended mixing in water. The aggregates formed by this method are heterogeneous in size (20 nm and larger) and shape (angular to round), but are crystalline in structure – exhibiting a face centered cubic (FCC) habit as determined by electron diffraction. In addition, particle shape and surface charge change when C₆₀ was mixed in the presence of electrolytes (NaCl, CaCl₂) or low molecular weight acids (e.g., citric, tartaric, acetic). A particular focus has been made on the characterization of the aggregates produced in the presence of these low molecular weight acids due to their environmental relevance. The aggregates formed in the presence of these acids tend to be more homogeneous in size than those mixed in water alone. In addition, the average diameter of the aggregates is smaller in the presence of low molecular weight acids than in their absence. The implications of these differences will be discussed in terms of both particle stability and how they affect C₆₀ transport through a sand-column.

Transport of colloidal tracers in a natural porous medium: impact of pore water velocity, size and zeta potential of nanoparticles on transport parameters and deposition rates.

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Groundwater is an important receiving environment for bulk chemicals of daily use and is therefore expected to be a sink for nanoparticles as well. Indeed, the mobility of a particle in a porous medium is strongly limited by its tendency to deposit on the surface of the grains, the so-called “filtration-effect”. The process of (nano)particle deposition during flow through porous media is commonly assumed to take place in two rate-limiting steps [1]: first transport to the surface of the collector by Brownian diffusion, interception, or sedimentation (for the larger ones), then, attachment to the surface. The kinetics of the transport step depends primarily on physical factors such as size, shape, density of the nanoparticles, flow velocity and pore geometry. The kinetics of the attachment step is controlled by interparticle forces: van der Waals, electrostatic, steric repulsion and hydrophobic forces between nanoparticles and grain surfaces. Due to the large number of factors influencing nanoparticles mobility in porous media, we propose here to use versatile model colloidal tracers, called “nanotracers” to gain insight into the impact of the mechanisms that drive the transport and the deposition of nanoparticles in a natural porous medium.

Monodisperse silica fluorescent nanoparticles were obtained from a modified Stöber synthesis [2]. The use of nanotracers with controlled properties allows quantification of the impact of size and the zeta potential on the mobility of the particles individually. Therefore, after production of functionalized nanotracers using silane coupling agents [3], and characterisation by Dynamic Light Scattering technique, leaching experiments were performed in repacked columns under conditions representative of groundwater flow. The soil used is a calcareous alluvial deposit from a fluvial shallow aquifer. The produced nanotracer suspensions have been injected into columns of 25-30 cm in length and fluorescence followed in the outflow to determine the breakthrough curves. These results have been described using the classical convection-dispersion equation, with a first-order irreversible process for particle deposition. Correlations between transport and deposition parameters using size and zeta potential of the nanotracers have been established under environmentally relevant conditions. The deposition rates have also been compared with predictions made using the DLVO theory and the classical filtration theory [4], that it is known to be hardly applicable for particles transport through natural porous media.

The results obtained indicate that the use of designed colloidal tracers is a promising tool for the estimation of nanoparticles transport parameters.

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Surface reactivity of nano-oxides and biological impacts

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The growing interest in nanotechnology is comparable to a new industrial revolution. The synthesis of functionalized nanoparticles (NPs) allows the development of new and innovative applications as imaging contrast agent, drug delivery, water treatment, catalysis, electronic, energy... In the environmental field, NPs provide solutions to several environmental issues due to their surface reactivity and large specific surface area. The surface reactivity of oxide NPs has been studied through the adsorption of a molecule probe as As(III). We have shown the strong retention efficiency of As ($8\text{As}/\text{nm}^2$) by iron oxide NPs (6nm). The adsorption site density dramatically increases when the size of the nano-oxides is <20 nm due to the presence of unusual adsorption sites and to the significant decrease of surface energy during adsorption. These sites are particularly sensitive to oxidation of the adsorbates. The surface speciation and the determination of the adsorption sites have been carried out by using X-ray absorption spectroscopies (EXAFS and XANES).

In biological media the reactivity of the oxides NPs has been ascertained through these methods allowing to characterize the redox activity at the surface when nano-oxides are in contact with biological models. Indeed, even if the amount of NPs produce by industry is limited the perspective of large worldwide production trigger off fears of peoples as well as scientists. Several properties confer to the NPs unique behaviors in contact with cells: their small size favors diffusive transfer and their reactive surface can lead to interaction with object with comparable size (membrane, proteins, DNA...). We have evaluated the potential cyto- and genotoxicity of oxide NPs towards human cells and bacteria to understand the mechanisms of interaction. A classification of the toxicity of oxide NPs as a function of their redox properties has been proposed. While chemically stable NPs ($\gamma\text{Fe}_2\text{O}_3$) in biological media have no apparent toxicity, NPs with a strong oxydant (CeO_2) or reductive (Fe°) power appear cytotoxic for *Escherichia coli* and genotoxic for human fibroblasts.

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Mobility of iron oxide nano particles in saturated porous media

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The use of engineered nano materials (ENM) is becoming common in many industrial and domestic applications and is expected to become widespread in the near future. Together with the great promise of this new group of materials a concern is raised regarding potential uncontrolled exposure to the ENM. Therefore methods for detection and analysis of ENM fate and behaviour in the subsurface environment are urgently required. Accordingly, three aspects of ENM fate in the subsurface environment are considered in the presented study. First, a new analytical technique to quantitatively measure concentrations of nanosized iron oxide particles in solution, based on UV-vis spectroscopy is presented; this technique complements optical measurements. We then demonstrate that sorption of these nanoparticles on various artificial and natural porous media is essentially negligible, within experimental error; the addition of humic acids to the porous media marginally increases sorption, but yields remain less than 5%. Finally, the transport and distribution of nanoparticles in water-saturated columns packed with porous media is examined. We find that transport behavior is influenced strongly by the method of nanoparticle introduction at the column inlet. Application of dry nanoparticles at the column inlet, followed by periodic flow of water, leads to highly limited nanoparticle migration. Use of continuous water flow causes the nanoparticles to undergo migration, settling and retention, with small amounts of elution from the column outlet. In contrast, nanoparticles that enter the column in a flowing suspension are transported through the column with virtually no retention effects.

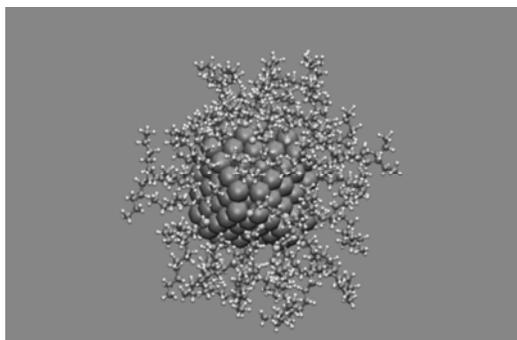
Model the Nanoscale with Chemical Accuracy: How Multiscale Modelling Simulations Can Help

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At the nano-scale, traditionally separate research fields such as chemistry, physics, and materials technology merge. As a consequence, computational materials science methods need to be developed in order to answer to new kind of questions.

Multiscale modelling approach, i.e. taking into account different time and length-scales in the computational model of nanostructured materials seems to be a strict requirement of any computational method aimed to give not only qualitative but also quantitative answers.



The presentation will be focused on different methods developed and implemented in the MoLNaC group with special attention on new class of methods named “ systematic coarse-graining” and “ reverse-mapping” applied to polymeric materials and recently extended to the study of nanoparticles. In the picture, a typical atomistic model of a gold Nanoparticle passivated by alkanthiolate is sketched.

Effect of natural aquatic colloids on manufactured nanoparticles (iron hydroxide nanoparticles and single-walled carbon nanotubes) in aquatic system

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There are wide applications of engineered nanoparticles (one dimension < 100 nm in size), including health and environmental benefits due to their distinctive properties (mechanical, thermal, optical, electrical, and magnetic). However there is also a need to understand the fate, behaviour and persistence of nanoparticles in environment, and their potential ecotoxicity. Iron hydroxide nanoparticles (primary particle ca 7 nm) and carboxylic acid functionalized single-walled carbon nanotubes (10 nm wide by 500 nm long) were analysed before and after their mixing with natural organic nanoparticles (Suwannee River humic acid, Suwannee River fulvic acid, polysaccharides and filtered natural waters) by a variety of techniques including electrophoretic mobility, DLS, HR-TEM and 3D EEM fluorescence spectroscopy as a function of interaction time, pH, organic nanoparticles concentration and temperature. The aim of the study was to monitor the influence of organic matter on the aggregation and precipitation processes.

Significant interactions of natural and engineered nanoparticles were observed including changes in nanoparticle charge, size distribution, aggregation status, and sedimentation behaviour. Evidence of thermal and metal fluorescence quenching of fulvic acid was found. These findings have important implications for the fate and behaviour of engineered nanoparticles in the environment and their ecotoxicity.

Interactions between natural aquatic colloids and manufactured nanoparticles: effects on chemistry, transport and ecotoxicology

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Manufactured nanoparticles (NPs) have been shown to be potentially toxic to aquatic organisms in the environment and are being produced, and likely discharged to the environment, in ever greater amounts. These NPs represent a significant pollution problem which will become more severe in future.

Once in the environment, NPs will interact with natural components such as natural organic and colloidal matter (humic substances, polysaccharides and other material) and be influenced by ambient solution conditions (pH, ionic strength and Ca concentration). All these factors will influence the physico-chemical behaviour and speciation of the NPs. In particular, dissolution, surface properties and aggregation will be altered. These changes will clearly influence their fate and behaviour in the environment.

This presentation will consider the interactions between natural colloids and NPs, along with other solution conditions, with reference to specific examples. In addition, the likely impact of toxicity and transport of these interactions will be considered.

On-line procedures for the analysis of nanoparticles in air

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A number of techniques to characterize nanometer sized particles have been developed in aerosol science during the last years. Many of them are based on the interaction of particles with electromagnetic radiation (mainly light) or interaction with the surrounding gas (diffusion, inertial effects, or attachment of labeled molecules as for example ions, charging particles electrically). This allows determining quantities like number concentration, size or mass. Combining several techniques additional information as density or particle structure as for example in terms of the fractal dimension can be obtained.

Optical techniques (light -scattering, -extinction, -absorption) have the great advantage that remote measurements are possible which is in particular useful if measurements have to be done in an aggressive environment. Multi angle or multi wavelength scattering also yields size and morphology information (Sorensen, 2001, Keller et al., 2006).

Once particles are charged electrically, they can be manipulated by electrical fields and detected by the electrical current, they cause. Based on electrical charging very compact, portable devices can be built. For example the Diffusion Size Classifier (DiSC, Fierz et al., 2008) yields particle number concentration and average size. Size information in this device is obtained by a one stage diffusion battery. If several stages are used, not only the average diameter, but also the size distribution can be determined. Charging particles by attachment of ions, produced in a corona discharge, is a mainly material independent process. If, on the other hand, particles are charged by emission of electrons upon irradiation with uv-light, also chemical information can be derived, for example particles arising from incomplete combustion can easily be distinguished from most other particles. Combined with thermodenuders (Fierz et al., 2007) a separation between volatile and solid material can be achieved.

Features, limits and applications of several techniques will be discussed.

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An advanced sampling device for improved analysis of fine and ultrafine atmospheric particles with synchrotron radiation

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Air pollution by particulate matter below 10 micrometer (PM10) is of public concern both in the area of legal regulations for PM10 reduction and nanotechnology risk assessments. A special focus is put on the trace elements present in PM10, because they show a potential for adverse health effects and are emitted into the atmosphere by industry and traffic in considerable amounts in addition to their natural abundance. Several elements also represent specific fingerprints for individual emission sources and are thus thoroughly investigated for source apportionment information. Many activities that influence the air pollution levels usually change within a few hours (for example rush hour traffic). Therefore time-resolved monitoring of trace elements in ambient air is highly important. This, however, requires novel devices for representative sampling and extremely sensitive analytical techniques.

A size-specific particulate matter sampler was constructed and optimized for subsequent trace element analysis. The Rotating Drum Impactor (RDI) collects ambient aerosol in three size ranges that are relevant for human exposure (2.5-10 μm , 1-2.5 μm and 0.1-1 μm) on aluminum sample holder wheels. The sampling time resolution is selectable, achieved by stepwise rotation of the aerosol collection wheels. The hourly aerosol samples show a barcode like deposition pattern. The total aerosol mass collected during an hourly sample interval is lower than 10 μg and contains trace elements in the picogram to nanogram mass range. Their detection requires special analytical techniques. Synchrotron generated X-ray radiation shows a very high photon flux density and is sharply focused. This, together with the non-scattering behavior of the substrate holder system, allows for the highly sensitive detection of trace elements in the rotating drum impactor samples. Absolute calibration of the measured XRF spectral data is performed by external calibration (TXRF, ICP-OES) of homogenous standard samples prepared with an ink jet printer. Synchrotron X-ray fluorescence spectrometry (SR-XRF) has been applied as a powerful fit-for-purpose tool for the detection of a broad range of trace elements (Bukowiecki et al., 2005). The average uncertainty of measurement for the elemental ambient concentrations is $\pm 20\%$. The unique set-up has been successfully applied for the characterization of railway emissions (Bukowiecki et al., 2007) and is now applied to study the contribution of road traffic abrasion particles to overall PM10 burdens. For the first time, hourly concentration levels of antimony in urban ambient air have been determined.

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3D Chemical Mapping of Nanoparticles in Cells

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In recent years, the concern over the rapidly increasing number of industrial, consumer, and biomedical products and manufacturing processes that utilize nanomaterials is driving government and industry efforts to develop accurate and reliable assessment methods for the health and environmental effects of nanomaterials¹. Many types of short term and long term nanoparticle exposure studies have been performed using cells and complex organisms. However the routes of exposure and the mechanisms of toxicity for many nanomaterials are still poorly understood^{2,3}. One of the biggest challenges in these studies is the accurate and reliable detection, localization, and quantification of nanoparticles that have been taken up by the cells and tissues.

We use focused ion beam scanning electron microscopy (FIB SEM) and energy dispersive x-ray spectroscopy (EDS) to examine the chemical and the morphological states of quantum dot exposed *T. pyriformis* in 3-dimensional (3D) space. Although it is possible to detect fluorescent nanoparticles such as quantum dots in these organisms using optical microscopy, the ability to resolve spatial details of intra-cellular localization patterns is limited. With the high resolution imaging capability of an SEM and the site specific nano-scale milling functionality of a FIB, we can produce high resolution 3D maps of the cellular landscape.

Since conventional SEM or TEM images rely on the atomic number differences in cellular components for contrast, it becomes difficult to identify and differentiate features with similar atomic masses. For example, small cellular debris with heavy metal staining might look very similar to quantum dot aggregates. Accurate quantification of nanoparticles requires unambiguous identification of nanoparticles among other cellular components. EDS mapping after each milling step produces a stack of elemental maps that can be processed into high resolution 3D chemical maps.

There are multiple techniques that can provide population-based or bulk morphological and chemical analysis of cells. However, the ability to detect and measure the intracellular chemical and morphological details at high resolution can provide valuable insight into the nanoparticle uptake and toxicity mechanisms.

FIB SEM EDS technique is still at an early stage of development and much work needs to be done. Although it is possible to perform qualitative 3D chemical analysis at high resolution, routine quantitative analysis of nanoparticles in biological systems at intracellular resolution will require development of reference maps and methods relevant for biological specimen.

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High-Resolution Measurement of Particle Transport in Variably-Saturated Porous Media using Time Lapse Fluorescence Imaging

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Colloidal particles and nanoparticles exist naturally in soil and groundwater and are present in contaminant streams entering the environment. The deposition, remobilisation and transport processes of the particulate phase are implicated in the leaching of nutrients from soils (McGechan and Lewis, 2002), enhanced movement of contaminants through the soil profile (Kretzschmar et al., 1999), and the efficiency and effectiveness of *in situ* bioremediation (Ginn et al., 2002) and wastewater filtration systems.

We report a set of laboratory visualisation and analytical results that use novel methodologies that enable high spatial and temporal resolution measurement of particle transport in quartz sand packs under a range of flow and transport conditions. The visualisation technique, Time Lapse Fluorescence Imaging (TLFI), uses ultraviolet (UV) light to excite visible-light fluorescence in dyed synthetic latex colloids (1.9 μm diameter) and an inert solute tracer passing through a thin bed of translucent sand. The intensity of fluorescence is directly proportional to the quantity of particles at any point in the pseudo-two dimensional system. Hence, variations in particle concentration at any instant in time can therefore be resolved at millimetre scale over a decimetre-scale domain (Bridge et al., 2006). We have also established a relationship between fluorescence intensity and pore water content (saturation) that enables the measurement of particle transport within unsaturated and variably-saturated quartz sand (Bridge et al., 2007).

We present the latest application of this technique to the problem of particle mobilisation and redistribution under changing saturation gradients during drainage of a sand pack. We find that the mass of particles remobilised and the mechanisms of attachment and detachment at the drying front are dependent on the ionic strength of the pore water during drainage. Furthermore, we present evidence that mobilisation kinetics are also influenced by the distribution and morphology of the immobile particles prior to drainage. This suggests that the remobilisation and transport of colloids in similar natural systems (e.g. soil profiles under drainage and infiltration cycles) may be sensitive both to initial and antecedent conditions at any given location. Our experimental data provide a new and quantitative insight into the nature of this coupling. The TLFI technique has potential for further application to functionalised particles of any size, including bacteria and nanoparticles.

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Highly sensitive detection of environmental pollutants by Laser-induced Breakdown Detection (NPA/LIBD)

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The Karlsruhe Research Center/Germany has developed a Nano-Particle Analyzer based on the Laser-induced Breakdown Detection method (NPA/LIBD); it has been applied successfully to various fields of environmental sciences. Key features of the technology are: Highly sensitive analytical tool for the determination of size, concentration and number density of nano-particles (colloids) in liquids [1, 2]. Compared to laser light scattering and obscuration the NPA/LIBD shows a significantly better detection limit by several orders of magnitude, especially for particles <100 nm. Limit of detection is a few ng/L for 10 nm colloids (typically 10⁵ particles/mL) and several mg/L for 1 µm particles (10⁴ particles/mL). The method is best suited for particles in the colloidal size range, i.e. from about 10 to 1000 nm, but the range can be extended to over 3 µm [3]. Typical measurement duration is several minutes, no sample preparation is required, and the analysis is largely non-invasive.

Colloids in the lower nm size range are the predominant species in all kinds of natural aquatic systems [4]; the same goes for technical systems, and it is the smallest of particles which are the hardest to remove. Due to their high surface to volume ratio they can adsorb significant amounts of pollutants which in turn opens up a way of transport over large distances even for otherwise insoluble substances [5]. They can further provide a medium for bacterial growth, and even bacteria themselves are often referred to as bio-colloids [6]. In industrial processes nano-particles are generally either unwanted impurities or the process product; both need to be detected and characterized.

Due to its unparalleled sensitivity the NPA/LIBD is a rather novel, valuable tool for studying all kinds of nano-particles without artefacts related to sample preparation. A combination with colloid fractionation techniques like field-flow fractionation further expands its possibilities. The NPA/LIBD closes the gap to established but more insensitive particle detection methods. One of the latest applications of NPA/LIBD has been in the field of drinking water purification by means of UF membranes. It has been shown for the first time that despite the membrane manufacturer's nominal cut-off of ≈ 10 nm colloids of up to 70 nm can be found in the permeate. First results and consequences are presented in our presentation.

So apart from enlarging the scope of current particle analysis technology the NPA/LIBD has the potential to play an important role in future key areas such as *online* monitoring of membrane integrity and *online* detection of hazardous biological material in water.

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Nanoparticles detected in the runoff of an urban area

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Engineered nanoparticles (NP) are used in a variety of products applied indoor and outdoor. Most popular applications include paintings, facades and tires. The amount of NP that are produced and applied in the various products can be assessed based on sales information. However, very little data regarding the release of NP to the environment is available. This gap in knowledge is caused by the lack of adequate analytical techniques to detect and characterize NP in the aquatic environment. Frequently used sizing techniques are based on light scattering and cannot discriminate between natural nanosized materials and engineered NP. In surface waters, the number concentration of naturally occurring nanosized materials is in the range of $10^8 - 10^{11}$ particles/mL. Engineered NP released (unintended) to the environment will be rapidly diluted in natural waters and therefore naturally occurring nanosized materials will overwhelm engineered NP in aquatic environments. Thus, an identification of engineered NP in the aquatic environment based on the number and the size alone seems impossible.

The TEM (transmission electron microscope) allows the detailed analysis of individual particles in the nanometer size range. Using the TEM (in combination with an elemental analysis such as EDX (energy dispersive spectroscopy) or EELS (electron energy loss spectroscopy) which is also performed in the TEM) the size, morphology, chemistry and crystallography of the particles can be determined. This information of particle characteristics allows a distinction between naturally occurring nanosized materials and engineered NP in the aquatic environment.

As a first case study, we have analyzed runoff waters from an urban area using TEM / EDX. For that purpose, 10 mL of runoff was collected and centrifuged (30 min, 3300 x g) directly on TEM grids. Results indicate that a high number concentration of TiO₂ NP is present in the runoff. TiO₂ occurs naturally in geological material (as rutile or anatase) and these particles could therefore also have a natural origin. However, based on the spherical morphology of the particles revealed in the TEM, a natural origin can be excluded and the TiO₂ particles can be assigned to an anthropogenic source (= engineered NP).

This example shows that a very careful and detailed analysis is required if natural nanosized materials are to be distinguished from engineered NP. In the case of TiO₂, neither size nor chemistry is sufficient to distinguish between natural and engineered particles. Only the additional information about the morphology as revealed by the TEM analysis allowed an assignment of the TiO₂ particles to an anthropogenic source.

In vitro assessment of nanomaterial toxicity: need for better characterisation of materials and methods

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The increasing production, particularly of metal oxide nanoparticles (NPs) and new carbon materials, will enhance the possible exposure at work places, packing stations and during application of the products. In addition, waste treatment and containment at the end of a products life cycle must be considered. Because of all these reasons, it is of great interest to determine how these materials, when coming in contact with living organisms, are taken up, transported in or through cell layers, and affecting biological functions.

To cover these questions a multitude of studies started during the past 5 years worldwide. Nevertheless missing definitions or standardised methods make most of these studies not usable/comparable. Thus, this short overview on biological hazards of nanomaterials will present a more general aspect of the possible risks that are connected with the production and use of nanomaterials and possible flaws which may reduce the benefit of many studies.

The increasing use of carbon nanotubes (CNTs) in various products underlines the need for a better understanding of their potential side effects. As they were produced by different methods as single-walled or multi-walled CNT they are generally contaminated by amorphous carbon and metal catalysts. Thus, the difficulty to assess CNT toxicity comes along with the material diversity but also with the diversity of the studies published so far. Within all our experimental set-ups we focused therefore on exact characterisation of the material to clarify various faults and misinterpretations from the literature.

We have found that well-dispersed CNT-bundles are less cytotoxic than rope-like CNT agglomerates in human cells, suggesting the role of agglomerates in the cytotoxic effect of CNTs [1]. Additionally, we present data indicating that CNTs are able to cross the cell membrane of human lung epithelial cells (A549) and may lead to adverse biological effects. Cells were incubated with non-purified, as well as amorphous carbon free and metal catalyst free acid treated single-walled carbon nanotubes. We do not observe any acute toxicity on cell viability as measured by WST assay and cell membrane integrity upon incubation with CNTs though these carbon conformations fake cyto-toxic effects in the MTT assay [2,3]. Furthermore, we observe an almost four fold increase on the release of reactive oxygen species (ROS) whereas incubation with contaminant free SWCNT have not such an effect [4,5].

Thus, our results suggest that

1. mostly the contaminants induce the acute toxicity of CNTs
2. analytical procedures must be carefully assessed to be applicable for these investigations
3. solvents to disperse NPs have to be carefully tested for their intrinsic effects in vitro
4. altered chemical behaviour of NPs must be taken into consideration (compared to bulk)
5. the morphology of the CNTs has to be characterised and described very well within publications as this is important for the interpretation of the results.

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Carbon Nanotube Fate and Reactivity Following Extended Simulated Groundwater Exposure

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Globally over the past several years, the escalating development, production and use of engineered nanoparticles in commercial products has presented serious questions about the disposal and fate of these materials in the environment. With the recent use of nanoparticles in various commercial products (i.e. tires, athletic equipment and clothing, paints, cosmetics, structural components etc), the possibility of environmental contamination through disposal via drains and landfills with resultant ground water contamination presents an immediate problem to not only adjacent biota, but directly to humans. This study examines changes in physical (morphological, charge and compositional) characteristics, and human lung cell response to freshly prepared, and 2.5 yr aged carbon nanotube (CNT) preparations. Two types of CNT preparations commonly used in commercial products were used in this study: (1) as prepared CarboLex (Carbolex, Inc., Lexington Kentucky) containing SWCNTs, nanoropes, graphene, graphite, Ni and trace Y; and (2) Air Oxidized SWCNTs (prepared from the same Carbolex) with trace Ni and no remaining Y. Both 10 uM and 100 uM CNT preparations were made in phosphate buffered saline pH 6.5-6.8, or fresh water, and for consistency, the same CNT batch was used to make all preparations. Each sample preparation was analyzed by TEM and X-ray microanalysis to monitor sample morphology and metal and salt content. Zeta analysis, UV-vis, and FT-IR were used to examine CNT charge, concentration and composition in the fresh and aqueous-aged preparations. Human NCI-H292 lung cell monolayers were grown to confluency in low serum medium and incubated with 10uM or 100uM CNT preparations for 3 hrs at 36°C with constant rotary swirling to disperse the nanoparticles during incubation. Following incubation the media was removed from each monolayer and saved for later analysis (to identify detached cells, contaminants and unattached nanoparticles), and the monolayers were either (a) stained with vital dye (erythrosin B in PBS) for viability analysis; (b) fixed in 3% glutaraldehyde in 0.1M cacodylate buffer with 10% sucrose followed by 1% OsO₄ fixation and prepared for TEM (thin sectioned), or (c) critical point dried and imaged by field emission SEM (JSM6500FE). Freshly prepared Carbolex and Air Oxidized CNT preparations caused plasma membrane damage to the monolayers where the CNTs attached to the apical cell surface causing small (50-90nm by TEM) electron lucent areas in the plasma membrane, as well as apical membrane tears and holes (seen by FESEM). Monolayer cells without attached CNTs grew normally and showed no morphological abnormalities by light or EM. In contrast, the 2.5 yr aged-aqueous preparations of Carbolex and Air oxidized CNTs did not produce severe damage to the cells, even though the aged preparations were CNT aggregates. Viability studies showed that fresh preparations of 100 uM Air Oxidized nanotubes produced a 3.8% increase in lung cell necrosis (compared to controls), whereas the PBS 2.5 yr aged 100uM Air Oxidized CNTs produced only 1.5% increase in cell necrosis. Similarly, freshly made 100uM Carbolex CNTs in PBS increased cell necrosis by 4.5%, whereas the 2.5 yr aged Carbolex 100uM increased necrosis by 1.6%. EM of the aqueous-aged CNT preparations showed various sized aggregates (20nm-0.5mm), however these aggregates were found to be less cytotoxic than the dispersed CNTs in the freshly made preparations.

Distribution and fate of nanodiamonds in vivo

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Recently, diamond nanocrystal, owing to its extreme chemical inertness, optical transparency, exceptional hardness and excellent biocompatibility, is highly expected to be a versatile valuable material in many areas such as biosensors, drug carriers, and imaging probes etc... Understanding the basic biological consequence of nanodiamonds is crucial to realize their applications and ascertain the possible hazards of nanodiamonds to humans. The aim of the present study is to reveal the biodistribution and translocation of nanodiamonds in mice, providing fundamental information for the biosafety assessment of nanodiamonds.

Well-characterized nanodiamonds with a diameter of around 50 nm were used in the experiments. Bovine serum albumin (BSA) was used as a dispersant to disperse nanodiamonds. Using ¹²⁵I labeled nanodiamonds as the tracer, the distribution of nanodiamonds with a dosage of 20 mg/kg b. w. in mice was investigated via single intravenous injection. It was found that nanodiamonds predominately accumulated in mouse liver; spleen and lung were also target organs for nanodiamonds. Whereas, no obvious accumulation was observed in the other organs. The changes of hepatic and lung distribution of nanodiamonds during 28 days post exposure were studied using spectroscopic method. Analysis of the digested solution of liver and lung shows that about 50% of initial nanodiamonds was entrapped in mouse liver at 3 h post dosing, and this datum kept constant within 28 days. Transmission electron micrographic imaging and Raman spectrum of the digested organ solutions confirm definitely the long entrapment of nanodiamonds in mouse liver.

Toxicity and photo-toxicity of nanoTiO₂: *in vitro* ESR and *single-cell-level* AFM assays

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NanoTiO₂ is the most popular semiconductor system used in heterogeneous photocatalysis and UV filtering materials. Here, we report on electron spin resonance (ESR) and atomic force microscopy (AFM) studies of the ability of nanoTiO₂ to generate reactive oxygen species (ROS), and thus the oxidative stress, *in vitro*. Commercially available nanoTiO₂ (anatase, primary particle size of ~6 and 20 nm) and custom-made TiO₂-based nanotubes (~60 nm in diameter and 1-5 μm in length), were used. In ESR experiments, aqueous suspensions of nanoTiO₂ were illuminated with UV-A light (λ=365 nm) and a spin-trap, DMPO, in combination with selected ROS-inhibitors, revealed generation of superoxide (O₂^{•-}) and hydroxyl (OH[•]) radicals. In the parallel study, the production of ROS by living cells (human neutrophils) exposed to the presence of nanoTiO₂ in the dark was checked by ESR. *In vitro* ESR revealed generation of both O₂^{•-} and OH[•] radicals and set the stage for *single-cell-level* AFM studies of the cellular stiffness in individual living human skin fibroblasts (CCL-110) and bladder cells (HCV29). The cells were exposed to nanoTiO₂-mediated photo-oxidative stress directly under the AFM tip. The AFM force-spectroscopy measurements were performed for cells exposed to the deleterious action of ROS generated by UV-A (8 and 20 mW/cm²) or UV-C light (0.1 mW/cm²) in aqueous media (PBS) containing concentrations of nanoTiO₂ in the range 0.05 - 0.5 μg/mL. The AFM measurements performed for cells illuminated with UV-A revealed a rapid drop (time scale < 1 min) of the cellular stiffness, E_{cell} , being of ~30-60%, which scaled with the exposure to the oxidative stress. These findings point to nanoTiO₂-mediated photo-oxidative stress on living cells, which results in reorganization of the cytoskeleton. They also highlight the sensitivity of AFM to detect early changes in mechanical properties of cells exposed to the photo-oxidative stress.

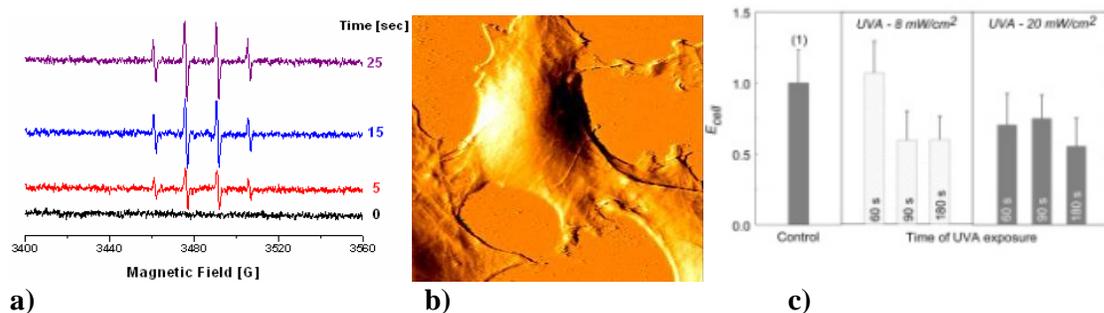


Figure 1. (a) Typical time-dependent evolution of ESR traces of DMPO-OH resulting from photogeneration of O₂^{•-} and OH[•] radicals in the presence of nanoTiO₂. (b) AFM image of the human fibroblast. (c) Relative changes in E_{cell} for fibroblasts exposed to UV-A (8 and 20 mW/cm²) in the presence of nanoTiO₂.

Microbial Interactions with Fullerene and Other Engineered Nanoparticles

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The rapid development of nanotechnology is outpacing the development of appropriate regulations to mitigate potential risks associated with their release to the environment. Therefore, we have been conducting research to improve our understanding of how fullerenes and other engineered nanoparticles interact with microorganisms, which form the basis of all known ecosystems. Buckminsterfullerene water suspensions (nC₆₀) exerted potent antimicrobial activity, similar to that of nano-silver. The antimicrobial activity of nano-sized ZnO, TiO₂, zero-valent iron and SiO₂ was significantly lower. nC₆₀ is a potent antibacterial agent when tested against pure cultures of different bacteria, and increasing exposure time and dose both increase toxicity¹. Toxicity was increased by smaller particle size in a manner disproportionate to the increase in surface area to volume ratio. nC₆₀ also retains its antibacterial activity for over 2 years, in the presence or absence of light, and under both aerobic and anaerobic conditions. However, toxicity is mitigated by salts which promote precipitation^{2,3}. While eukaryotic cell damage by fullerenes has been attributed to reactive oxygen species (ROS)⁵, we find no evidence of ROS-mediated damage in bacteria killed by nC₆₀⁶. Rather, flow cytometry studies showed that nC₆₀ acts as a direct oxidant that interferes with energy transduction. Furthermore, the methods used to evaluate ROS production and damage were confounded by interactions between nC₆₀ and the reagents, revealing a need to re-evaluate previous studies⁷. We also observed that the presence of geosorbents reduced the bioavailability of nC₆₀ and thus its antibacterial activity⁴. The adsorption of humic acid onto nC₆₀ also eliminated nC₆₀ toxicity, probably due to coating of the nC₆₀ and hindering direct contact with bacteria. This indicates the toxicity of nC₆₀ is controlled by its bioavailability in natural soil settings. Overall, this research reflects an overall image of preventable or negligible environmental impact of nC₆₀, and provides a methodology by which the potential environmental impacts of other nanomaterials can be evaluated.

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Ecotoxicology of engineered nanoparticles

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Engineered nanomaterials are exciting for industrial and consumer product development because they have different properties than the substances in bulk form. Current production volumes, combined with the envisioned growth in this field, leave no doubt that engineered nanomaterials will make their way into the environment. The question therefore arises as to if engineered nanomaterials pose a risk to environmental health and how this potential risk can be assessed. Various studies have recently appeared in the literature focussing, aside from bacteria, on hazard assessment in eukaryotes such as algae, plants, invertebrates and fish. Effects reported include, for example, reduced root growth in seedlings of certain plants due to alumina particles [1]; increased life-cycle mortality and reduced fertilization rates in the estuarine copepod *Amphiascus tenuiremis* due to non-purified single-walled carbon nanotubes [2] and gill injury and oxidative stress by titanium dioxide to rainbow trout (*Oncorhynchus mykiss*) [3]. Aside from these observations, two other interesting mechanistic aspects with regard to environmental impact of nanomaterials emerge. The first is the ability of organisms to alter the toxicity of nanomaterials as illustrated by an increased solubility of silver ions from silver nanoparticles upon interaction with algae [4]; the second is the influence of nanomaterials on the toxicity of common environmental contaminants [5]. The presentation will provide an overview of these reports and discovered phenomena and identify important next steps for linking effects with exposure.

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Ecotoxicology of nanoparticles Learning and relating to human toxicology

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Nanotechnology has resulted in the production of a vast array of nanoparticles that vary in many physicochemical characteristics, including chemistry, size, shape, charge and crystal structure. Many of these characteristics result in the unique properties that enable the nanoparticles to be exploited in many exciting applications. However, these properties are also likely to influence how these particles behave in the environment and in organisms. Some of these materials have been designed for applications that purposefully involve exposure to humans (e.g. suntan lotions and medicines), or to the environment (e.g. water filtration devices and zero valent iron for remediation). For such applications it is therefore essential that an understanding of their biological compatibility or hazard is attained before widespread use. For other applications (e.g. inclusion in aerosols, paints, IT products, food packaging and clothing), potential for exposure to humans and the environment of nanoparticles is also possible during manufacture, use or disposal of the product. The nature of such incidental exposures and any resultant hazard is currently poorly understood, but may be just as important as for purposeful exposures.

Due to the vast array of nanoparticles, formulations, uses and routes of release into the environment, it is impossible to assess the hazard associated with all potential exposure scenarios. For this reason strategic research priorities need to be identified that will lead to improved understanding. This presentation will summarise some of our understanding of the human toxicology of nanoparticles including cellular and molecular mechanisms of toxicity as well as physicochemical factors that appear to be important in driving toxicity. This presentation will propose integrating such knowledge and techniques into ecotoxicology studies in order to inform study design in order to accelerate our understanding.

Implications of Black Carbon Nanoparticles in the environment.

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The emission of black carbon is known to cause major environmental problems. Black carbon (BC) particles contribute to global warming, carry carcinogenic compounds and cause serious health risks. The characteristics of black carbon particles vary widely, but the smaller particles fall within the nanometer range. This means that part of the BC continuum falls within the nanoparticle size range. Extremely efficient sorption to BC carbon pulls highly toxic polycyclic aromatic hydrocarbons, polychlorinated biphenyls, dioxins, polybrominated diphenylethers and pesticides into sediments and soils^{1,2}. Therefore, black carbon nanoparticles (BCNP) may strongly reduce the risk posed by organic contaminants in sediments and soils. This increased sorption is general, but strongest for planar (most toxic) compounds at environmentally relevant, low aqueous concentrations. The presence of BC particles may reduce uptake in organisms by up to two orders of magnitude^{2,3}. Extremely strong sorption of hydrophobic chemicals also has been reported for carbon nanotubes (CNT)⁴ and one question is to what extent this can be compared to BCNP sorption in terms of sorption mechanism and sorption affinity. Another question is to what extent these nanoparticle – toxicant interactions are relevant for the total toxicity and risk assessment of NPs in the natural environment. It can be argued that the bioavailability of NP-bound toxicants is highly variable dependent on environmental conditions, as well as on the specific properties of these NPs, toxicants and organisms.

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Investigation of toxic potency of cobalt-doped tungsten carbide nanoparticles to rainbow trout gill cells (RTgill-W1)

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The production of nanomaterials increases world wide. Many applications already contain nanoparticles and new applications arise constantly. Even though production often takes place in closed systems, environmental exposure by spilling, leakage and waste disposal is likely to occur. In order to study the potential impact of released nanoparticles on environmental organisms, the rainbow trout gill cell line, RTgill-W1, was chosen as model. Cells were exposed to nano-sized tungsten carbide with or without cobalt doping. Both nanomaterials are used in heavy metal industry for extremely hard tools. Uptake und localisation was assessed by SEM coupled with EDX elemental analysis. Impact of nanoparticles on cell viability was assessed by fluorescence based cytotoxicity tests. To better meet environmental conditions, minimal culture media, which are closer in their composition to surface water than complete media, were tested as well to study the influence of media components on toxicity. Further, the influence of cobalt ions, a component of tungsten carbide cobalt which could potentially leak from the particles, was assessed in different media.

Results from the electron microscopic studies showed an uptake of both nanomaterials into RTgill-W1 cells, where localisation seems to be restricted to the cytoplasm, as no particles were observed in the nucleus of cells. Viability of RTgill-W1 cells was significantly reduced after exposure to both materials, but tungsten carbide cobalt caused a stronger effect than tungsten carbide alone. This effect was not due to diluted cobalt ions as shown by experiments with cobalt chloride. Exposure of RTgill-W1 cells to nanoparticles and cobalt chloride in reduced culture media did not influence particle effect, but enhanced the toxicity of cobalt ions.

These results indicate a hazardous potential of released nanomaterials to aquatic organisms.

Ecotoxicology of nanomaterials of differing structure: using data from the organism to the genome to elucidate the impact of nanoparticles on aquatic species

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There is a question as to how to predict the impact of exposure to nanomaterials on organisms, and specifically how small changes in the structure of a particle will impact the physiological response of an organism to exposures. Our lab is using two models, *Daphnia* spp. and trout (*Oncorhynchus mykiss*) to examine the impacts of nanoparticle exposures on immune function, behavior, mortality and genomics of aquatic species. This presentation will summarize several studies conducted in our lab investigating the impact of fullerenes and carbon nanotubes with different side chains and solubilities, as well as titanium dioxide, and gold nanomaterials on these two species. Toxicity, behavior and gene expression pattern differ with particle type and toxicity for each organism. In *Daphnia* we have determined that core particle structure has a significant impact on toxicity and effect on behavior. Side chains can mitigate this impact and may be a method for decreasing toxicity. Genomic data from both immunological tissues in fish and in whole *Daphnia* indicate that small changes in side chain chemical structure of a particle will affect the impact on physiology and immunological function. In addition, side chain structures appear to have a significant impact on toxicity that may surpass the particle itself in some cases. From this data we can summarize what characteristics of a particle may be the greater cause of toxicity so we can begin to make predictions about other types of particles to better inform risk assessment.

Toxicity and sorption of silver nanoparticles to *Chlamydomonas reinhardtii*

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Increasing uses of silver nanoparticles (AgNP) in a variety of industrial and consumer products makes their entrance into aquatic environments predictable. As a first step towards an ecotoxicological evaluation of AgNP we have examined for effects and sorption of AgNP and ionic silver (Ag^+) to the freshwater algae *Chlamydomonas reinhardtii*.

AgNP toxicity was hypothesized to relate to the uptake of ionic silver released from particles in the exposure medium or at the interface between AgNP and the algae. Examination of AgNP by TEM and Zeta Sizer showed that particles maintained their size distribution (ranging from 10 to 200 nm with an average of 40 nm) in the experimental media. Release of Ag^+ from particles was examined by DGT and ICP-MS and found to correspond to about 1 %. Toxicity to photosynthesis, determined by fluorimetry using a portable Pulse Amplitude Modulation Fluorimeter, was assessed to be higher for Ag^+ than for AgNP (177 and 2722 nM EC_{50} , respectively). Experiments carried out in presence of cysteine as strong silver ligand provided evidence for the role of ionic silver released from particles and the significance of direct interaction between AgNP and cells to induce toxicity. Preliminary results also indicate sorption of AgNP to algae. Examinations by TEM and SEM will provide information on the ultrastructural distribution of particles in algae.

Effect of micron- and nanosized titanium dioxide on algal growth: inherent inhibitory effects and as modifying factor on cadmium toxicity

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The effect of micron and nanosized TiO₂ on the growth of freshwater green algae *Pseudokirchneriella subcapitata* was studied in a series of algal growth inhibition tests.

It was found that effects of TiO₂ varied according to particle size, with smaller particles resulting in larger inhibition of algal growth. Furthermore the effects were found to depend on lighting conditions as lower effect concentrations were observed at lower light intensities.

Additionally the influence of the presence of TiO₂ nanoparticles on the toxicity of cadmium to *P. subcapitata* was studied through growth inhibition tests as well as accumulation studies.

Preliminary studies showed that addition of 2 mg/L TiO₂ changed the dose-response relationship for Cd(II) toxicity to *P. subcapitata*. By combining results from geochemical modelling, algal growth inhibition tests, and sorption studies it was shown that changes in toxicity to *P. subcapitata* varied according to the sorption behaviour of Cd(II) onto TiO₂. Fast sorption was observed with a sorption pattern of cadmium onto TiO₂ following a Langmuir adsorption isotherm. Similar experiments with crustacea *Daphnia magna* as test species was also carried out and results will be presented.

In accordance with the sorption studies and the observed reduced toxicity in the growth inhibition tests, exposure of algae to 2 mg/l TiO₂ in the presence of 40 µg/L Cd(II) for 2 and 24 hours resulted in a reduction in intracellular cadmium compared to experiments without TiO₂ added.

This study shows that that changes in the toxicity and uptake of cadmium in algae in the presence of TiO₂ may be explained by reduced bioavailability, due to a surface complexation reaction of TiO₂ with the positively charged cation Cd²⁺, combined with the inherent inhibitory effect of TiO₂ nanoparticles. Thus, the study underlines the importance of combining ecotoxicological testing with sorption studies to understand bioavailability and toxicity mechanisms in interactions between nanoparticles and chemical compounds.

Ecological Uptake and Depuration of Carbon-14 Nanotubes

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Carbon nanotubes represent one of the highlights to emerge from nanotechnology to date. Despite extensive research on their material properties and intended applications, the risks they pose to the welfare of humankind and the environment are not well understood. To characterize these risks, we have developed a technique for producing C¹⁴-labeled single- and multi-walled nanotubes, a technique that allows for direct quantification of unmodified nanotubes in biological and environmental samples. This approach overcomes many of the limitations of current techniques with regards to quantifying modified or unmodified single- or multi-walled carbon nanotubes. These nanotubes were purified and extensively characterized to ensure high purity with regard to the ratio of carbon nanotubes to amorphous carbon and also to catalyst impurities.

These radioactively labeled nanotubes were then sonicated and their uptake and depuration behaviors measured using the oligochaete (*Lumbriculus variegatus*), the earthworm (*Eisenia foetida*), and *Daphnia magna*. Carbon nanotubes were not shown to bioaccumulate in either *L. variegatus* or *E. foetida*, and nanotubes detected in these organisms appeared to be located in the guts of the organisms rather than absorbed into their tissues. Bioaccumulation factor values for the carbon nanotubes were at least an order or magnitude smaller than those for pyrene for both organisms. Uptake experiments were then pursued with *D. magna* to determine the extent to which carbon nanotubes are able to transport across the gut or dermal tissues to enter systemic circulation in the organisms. Unlike in the exposure experiments with *L. variegatus* and *E. foetida*, the nanotubes were not spiked to soils or sediments for the tests with *D. magna*. As such, these experiments help elucidate the extent to which nanotubes interactions with soils or sediments may influence their availability to ecological receptors.

The effects of dietary exposed nanosized titanium dioxide on terrestrial isopods

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According to the predicted future use of various nanoparticles, including titanium dioxide (TiO₂), they will most probably enter terrestrial environment. The inclusion of a set of terrestrial toxicity tests in risk characterization of nanoparticles is therefore necessary. At the moment, there is a lack of data on the effects of nanoparticles on terrestrial organisms and particularly their effects after oral uptake. We would like to present our work on the short-term effects of dietary exposed nanosized TiO₂ on terrestrial isopods *Porcellio scaber* (Isopoda, Crustacea). The isopods were exposed to nanosized TiO₂ (anatase, 15 nm) according to the protocol previously elaborated for dissolved chemicals (Drobne and Hopkin, 1994). The nanoparticles were applied onto leaves, which were offered to the animals. During the test, we monitored the changes on higher levels of biological organization, including the amount of ingested food, the defecation rate, the food assimilation efficiency, weight change and mortality. After three days of exposure, the isopods were dissected and the digestive glands were further analysed for enzyme analyses. Based on the reports that nanosized TiO₂ induces the production of reactive oxygen species (Rahman et al., 1997), we measured the activities of two antioxidant enzymes catalase (CAT) and glutathione S-transferase (GST). The activities of CAT and GST were decreased in a dose-independent manner. Namely, only the exposure concentrations 0.5 µg, 2000 µg and 3000 µg of TiO₂/g food decreased CAT and GST activities, but the intermediate concentrations (0.1; 1; 10; 100; 1000 µg/g food) did not result in significant changes of enzyme activities. Higher level responses were not affected up to the highest tested concentration of TiO₂ in the food (3000 µg/g food). Based on our experience, the test with terrestrial isopods is suitable for testing the effects of ingested nanoparticles, but the presentation of toxicity data needs to be adapted according to the mode of action of nanoparticles and their specific characteristics.

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Ecotoxicity of silica nanoparticles to the green alga *Pseudokirchneriella subcapitata*: importance of surface area

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One of the central hypotheses in nanotoxicology is that the toxicity of nanoparticles (NPs) is related to their large specific surface area. In the field of inhalation toxicology, it has already been shown that smaller NPs are more toxic than larger ones, and that the difference in toxicity can be attributed to the difference in specific surface area [1]. To date, this hypothesis has not been tested in ecotoxicology. The aim of this study was thus to investigate experimentally if the ecotoxicity of nanoparticles is related to surface area and not to mass.

Therefore, two commercially available silica (SiO₂) NP with different sizes were chosen for toxicity testing, i.e. LUDOX[®] LS and LUDOX[®] TM40 [2]. The experimental approach consisted of three parts: a physicochemical characterization of the NPs in standard OECD test medium [3], an assessment of the ecotoxicity effects of the NPs to a freshwater green alga and a transmission electron microscopy study (TEM) of the interaction between NPs and algal cells. The ecotoxicity assessment consisted of 72 hour chronic growth inhibition experiments with the green alga *Pseudokirchneriella subcapitata*. To assess the difference in toxicity between SiO₂ NPs with two different sizes, the NPs were tested simultaneously and this experiment was repeated 3 times more.

The silica NPs were stable in OECD test medium, with an average hydrodynamic diameter of 12.5 for LUDOX[®] LS and 27.0 nm for LUDOX[®] TM40. Colorimetric analysis of reactive silica in the test suspensions showed that the silica NPs did not dissolve enough to contribute to the toxicity response of the algae, meaning that the observed toxicity, as described below, was fully due to the interaction between NP and the algae. The EC20's (i.e., concentrations resulting in 20% growth inhibition) were (mean ± standard deviation, n=4) 18.2 ± 3.6 for the smallest NP (12.5 nm) and 27.9 ± 3.0 mg.l⁻¹ for the largest NP (27.0 nm). A t-test for dependent samples (n=4) indicated that the EC20 of the large NP was significantly higher than the EC20 of the small NP. However, when the EC20 was expressed as surface area concentration, a significant difference was no longer observed between the EC20 values of both NPs, with EC20 values of 4.3 ± 0.8 m².l⁻¹ for the small NP and 3.8 ± 0.4 m².l⁻¹ for the large NP. Hence, we conclude that the central hypothesis for the silica NP's investigated is valid and that the ecotoxicity of silica NPs is indeed related to surface area and not to mass. Further research with other NPs and test organisms (with different composition and a wider size range) is needed to validate this important finding.

Finally, a TEM study of algal cells exposed to toxic concentrations of both silica NPs did not reveal any uptake of nanoparticles into the algal cells. Rather, we observed that the NP's were adsorbed to the outer cell wall of the algae. This may suggest that the mechanism of silica NP toxicity to algae could be related to interactions with the cell wall. Here too, further research is required to unravel the exact mechanism of NP toxicity to algae cells.

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**The known “knowns” and known “unknowns”:
Mapping uncertainty in regard to the potential human and environmental
health risks of manufactured nanoparticles**

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As the number of products containing nanoscale materials continues to rise, questions still remain on the potential risks associated with their use and exposure in both our homes and environment. There are already over 500 nanotechnology-based consumer products on the market, while at the same time the scientific knowledge on the risks associated with nanoscale materials is still in its infancy. Although uncertainty and severe knowledge gaps surrounding potential negative impacts from exposure to nanomaterials are apparent and a corresponding understanding of the nature of uncertainty is fundamental for risk assessors and regulators, not much work has been done on this thus far. In this study we systematically map the scientific uncertainty by locating the areas of uncertainty through an in-depth analysis of governmental reports and scientific reviews dealing with the human health and environmental risks of nanomaterials. Once the locations of uncertainty were identified, we estimated the level and the sensitivity of the uncertainty.

The study shows that significant knowledge gaps exist not only in terms of documenting potential (eco)toxicological effects, but also in terms of characterizing exposure and nanoparticles behaviour even in simple test systems. For example, uncertainty related to testing strategies and environmentally realistic exposure scenarios, including establishing, developing and standardising reference materials, monitoring and detection equipment and estimating human and environmental exposure concentrations, impedes a successful risk characterisation of engineered nanoparticles according to several reports. These issues ultimately lead to significant challenges in performing human and environmental risk assessments and present a daunting task for regulators. We recommend that increased efforts are made by risk assessors and regulators to estimate the sensitivity of these knowledge gaps while simultaneously ensuring that the “right” scientific questions are addressed in order to effectively prioritise resources to reduce uncertainty most pertinent to an accelerated risk analysis of nanomaterials.

Greening Nanotechnology – new challenge or business as usual?

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In this paper I am interested in the parallels between the nanodiscourse and the sustainability discourse. A common and rather pessimistic reading is that both of them are involved in a rhetorical or even cynical game becoming the best and fastest growing economy in the global race supposedly for welfare and peace. In contrast to, I would like to suggest to take the challenges seriously being phrased in both discourses. They do not only seem to share a sense of the necessity - and also possibility - of monitoring and eventually controlling input and output of the production system: in short the dream of wasteless production. What's more, both of them refer to it in the sense of a moral obligation to avoid waste and also limit excess. It might thus be the effort worth to grant some credibility to the activities under the heading of a sustainable and "green" nanotechnology. This might be viewed as a success of readjustment of values in science policy making, an agenda that includes conducting research and educational projects that relate nanotechnology to social and environmental problems and that build bridges between nanoscientists/ engineers and environmentalists.

An analysis of the meanings and practices of space in both discourses lines up the whole discussion. The spatial conceptualization of nanotechnology makes it a sensitive field of technological and societal expectations, visions, but also of innovation. Looking at the spatial implications might become also a sound framing for nano-products, „to get grounded“ fears and high expectations. It can help to disentangle seemingly necessities. Turning Nano to Eco-Nano would then imply in first place turning nano from a global to a local framing, from a total, all-embracing nanospace to local eco-nanotechnologies.

Questions might then be for instance: What does "green nano" mean exactly, if we take road maps and policy advices serious? What does the greening of nano imply for dealing with concerns about nanotechnology products and toxicology? And is Ecologizing different form greening? And finally what are the philosophical and methodological lessons to be drawn from an eco-perspective?

US Research on Nanotechnology Applications: Green Nanotechnology for Past, Present, and Preventing Future Problems

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Green Nanotechnology is a responsible way to design and use nanomaterials and nanoproducts without harming the environment or human health. It involves both the production of nanomaterials and products using principles of green chemistry and green engineering and the designing of nanoproducts that provide solutions to environmental challenges.

In this paper I will present an overview of what is meant by green nanotechnology and some of the principles that guide its development, including a discussion of what might determine whether a product or process is really green. I will briefly cite research examples of using green nanotechnology to clean up past environmental problems such as hazardous waste sites and then address present environmental problems in which nanotechnology can be used, such as drinking water treatment, wastewater or end of pipe treatment, use of catalysts in present chemical processes, renewable nanomaterials in composites.

Designing nanomaterial processes to prevent future environmental problems will be discussed, highlighting processes such as using non-toxic solvents, self-assembling nanomaterials, solid state processes, DNA templating, bio-inspired synthesis, microwave techniques, photochemical synthesis, and others. Finally, nanoenabled green energy systems--solar, thermoelectric, fuel cells and hydrogen--will be included as another group of nanoscale processes which help prevent future environmental problems.

Ethical Reflections on Framing the Discourse on the Benefits and Risks of Nano-Materials for the Environment

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Society has come to expect socially responsive and responsible science. In the emerging nanoscale sciences a significant responsibility that is much discussed is the issue of whether or what risks are associated with manmade nanoparticles for humans, animals and the environment, and how should responsibilities be attributed for defining, identifying, managing, avoiding or keeping risks at an 'acceptable' level. Who should define acceptability; acceptable to who, based on what criteria, taking account of what benefits? What indeed are risks; what are benefits? The mainstream understanding of risk is that it can be calculated by using a formula such as: exposure + vulnerability + consequences, multiplied by probability. However is such an understanding of risk as being a logical and measurable dimension appropriate? Risk is increasingly acknowledged as being a complex concept that includes the rationally measurable as well as inherently subjective elements in the manner that risk is perceived and therefore assessed. Risk perception and assessment are a mixture of scientific judgment and psychological, emotional, social, cultural, and political factors. Not only are the views of the general public influenced by worldviews, ideologies, and values, so are those of scientists, particularly when they are working at the limits of their expertise. Different risks will furthermore be differently perceived by the various stakeholders: the scientists, the general public, investors, politicians, NGOs, regulators and ethicists. The complexity of the issue of how procedurally and substantively to decide whether and what risk are created by manmade nanoparticles for the environment, and which risks should be seen as being outweighed and justified by benefits, can be addressed by looking to the sustainable development discourse (that takes a systemic approach), combined with methods of applied ethics analysis. The three complex dimensions of sustainable development defined at the UN 1992 Rio Conference: environment, economy and society, are seen as being strongly and complexly interconnected i.e. actions and decisions in each area will affect the others. Applying a systems approach involves placing as much emphasis on the interface of the dimensions as on identifying and describing the objects and events themselves. Ethically the sustainability concept includes strong aspects of global justice and responsibility towards contemporary and future generations, and acknowledging certain duties towards the natural environment. This paper holds that in addition to (as noted in the conference Call for Abstracts), the assessing of the benefits and risks requiring a better understanding of their chemistry, mobility, bioavailability, and ecotoxicity in the environment, other inter- and transdisciplinary methodologies must be involved in order to fully assess the benefits and risks of nanomaterials in products and applications in a global societal and environmental context.

Polyelectrolyte Surface Modifications Optimize Emplacement and Mitigate Ecological Risk of Reactive Nanoparticles for In Situ Groundwater Remediation

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Reactive nanomaterials (e.g. nano-sized Fe⁰) are used for in situ remediation of contaminated groundwater (1-3). To be effective they must be reactive with the target contaminant and be mobile to some degree in the subsurface to enable emplacement at high particle concentrations. Enhanced mobility can be achieved through surface modification with polyelectrolytes (e.g. polyaspartate) or surfactants (e.g. SDS) however, surface coatings can also affect reactivity and toxicity (4-6). Concern over potential ecological risks (exposure and toxicity) of engineered nanomaterials introduced into the environment is motivation to determine how the properties of the adsorbed polyelectrolyte layer used to stabilize nanomaterial dispersions and various hydrogeochemical parameters (e.g. pH, ionic strength, flow velocity, grain size distribution) affect nanomaterial transport, reactivity, and toxicity. Understanding these relationships will enable design of surface modifiers to enhance emplacement in the subsurface while mitigating unwanted exposures and/or toxicity. This is one step towards sustainable development and implementation of nanotechnologies. Relationships between the adsorbed polyelectrolyte layer properties and particle aggregation (7,8) and deposition in water-saturated sand columns at low particle concentrations are described. The response of different surface modifiers to changes in ionic strength, pH, excess free polymer, and the presence of clays and fines is determined for low particle concentration where aggregation is low, and for high particle concentration where aggregation is significant. Mechanisms for decreased particle reactivity due to adsorbed polyelectrolytes are also presented. Finally, preliminary data on the effect of surface coatings on nanoparticle toxicity are presented. These studies emphasize the important role of geochemistry and aggregation on the transport of concentrated nanoparticle dispersions in porous media and highlight the tradeoffs between mobility, reactivity, and toxicity for different types of surface coatings.

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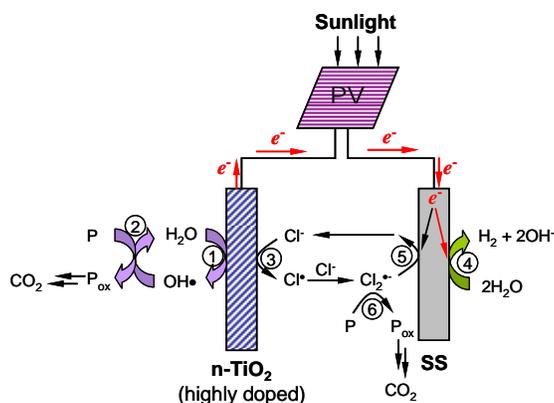
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Environmental Applications of Semiconductor Nanoparticles and Related Materials

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Nanoparticulate materials have been synthesized for more 100 years. During early stages of research they were explored primarily as a scientific curiosity in terms of their physicochemical properties, aggregation tendencies in colloidal suspensions, photochemical and catalytic properties, non-linear optical effects, and interfacial chemistry. At the present time, nanometer-sized semi-conducting particle arrays are used for the protective coating of a wide-variety of surfaces, for their self-cleaning properties when coated on glass, and for other diverse applications as environmental remediation agents, for the photo-splitting of water to produce hydrogen and oxygen, as semiconductor electrodes, as photo-initiator for polymerization reactions, and non-linear optical switches. In this keynote lecture, an overview of some of the unique properties and applications of semiconducting metal oxides, mixed-metal oxides, metal sulfides, and hybrid nanocomposites will be presented. In addition, a review of the quantum-sizing effects on particle reactivity with decreasing size will be presented¹⁻⁶.



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Plant-mediated synthesis of gold nanoparticles and their catalytic function

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Recently, the exploitation of biological systems has emerged as a novel technique for the synthesis of nanoparticles. There is a growing need to develop clean, non-toxic and environment friendly procedures ('green chemistry') for synthesis and assembly of nanoparticles, and thus the fusion of biotechnology and material science has heralded a frontier of nanobiotechnology. A perennial shrub, *Sesbania drummondii* removes potassium tetrachloroaurate (gold III ions) from the solution, and transforms gold (III) ions into a large quantity of gold nanoparticles in its tissues. Transmission electron microscopy revealed the intracellular distribution of monodisperse nanospheres, possibly due to reduction of the metal ions in cells. X-ray absorption near-edge structure and extended X-ray absorption fine structure demonstrated a high degree of efficiency for the biotransformation of Au(III) into Au(0) by plant tissues. We also demonstrate a catalytic function of the *in situ* generated gold nanoparticles while housed naturally in the plant tissue.

Degradation of persistent organic water pollutants using oxidation agents and copper oxide catalysts

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The continuous release of persistent organic chemicals such as pesticides, halogenated organic solvents, PAHs, and PCBs to aqueous surface and subsurface environments is an unfortunate reality. These compounds are recognized as toxic, and often carcinogenic and/or mutagenic, and they thus require highly efficient treatment procedures in aqueous systems. The current study presents an oxidation process to decontaminate polluted water, using nanosized copper oxide particles as catalysts, and hydrogen peroxide or ozone as oxidation agents. The process shows complete and rapid degradation of a wide range of organic contaminants under ambient pressure and temperature, in experiments lasting up to 30 minutes. In contrast, control runs measuring degradation through exposure only to hydrogen peroxide, or only to copper oxide nanoparticles, consistently show less than 10% reduction in contaminant concentration. Lack of exposure to light, and the particular method of mixing, appear to have no influence on the reaction rate or products. The reaction was found to proceed effectively in the range pH 3-8.5, and much slower at pH 10. Testing various concentrations of oxidation agent, an optimum point was found, with an increase above this concentration resulting in a reduced reaction rate. Moreover, measurements of reaction kinetics demonstrated conversion from exponential decay of a contaminant, typical of a first-order reaction, to a linear decrease in contaminant concentration, typical of a pseudo-zero-order reaction, upon increasing the oxidation agent concentration above a certain threshold. The copper oxide nanoparticles were characterized before and after the reaction, and also shown to retain reactivity over several cycles after refreshing the contaminant solution and adding more hydrogen peroxide. The turnover number for the copper oxide nanoparticles was ~ 5.3 molecules $\text{min}^{-1} \text{nm}^{-2}$.

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