Examination and assessment of consequences for industry, consumers, human health and the environment of possible options for changing the REACH requirements for nanomaterials

REFERENCE: IHCP/2011/I/05/27/OC

ANNEX 3 TO FINAL REPORT

IMPACTS ON HEALTH AND THE ENVIRONMENT IN DETAIL

09 January 2013

BiPRO
Beratungsgesellschaft für integrierte Problemlösungen

in cooperation with

Öko-Institut e.V.
Institut für angewandte Ökologie
Institute for Applied Ecology
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1 Introduction

Annex 3 elaborates human health and environmental impacts of 9 of the 21 options as proposed in the Nano Support Project 2012. The other 12 options are implicitly part of the current REACH regulation and the guidelines issued by the ECHA (see also baseline definition in the main document).

While this Annex necessarily focuses on the hazards and information needs connected to human health and the environment it is important to keep in mind, that nanomaterials are exceptionally useful and hold the promise to be a significant part of the solution to several pressing problems such the need to lower $\text{CO}_2$ emissions, more efficient use of natural resources etc. In connection with a balanced approach where research and development of new applications are accompanied by diligent and detailed risk assessment nanotechnology should become widely accepted in the population.

Three nanomaterials were selected as case studies to highlight the impacts the proposed options have in dependency of the toxicity already recognized from their bulk form, the tonnage bands and the pre-existing knowledge. Knowledge gaps resulting from failing to apply individual options arise in dependence tonnage bands and prior knowledge on the nanomaterial in question.

Additionally seven nanomaterials are discussed in broader terms without addressing individual options within their context. The application of endpoint specific options is very dependent on the tonnage band in which the individual materials are produced/imported. This presents a problem not only with regard to nanosafety (it has been proposed by authorities among them the German BAuA, UBA, BfR and the SRU to alter tonnage boundaries for nanomaterial risk assessment requirements) but also because one of the principal options (option 2) highlights an undecided issue concerning the treatment of nanomaterial surface treatment/modification as either a characteriser or an identifier (for elaboration see Annex I – Option Profiles). This issue remains without definite answer since the RIP-oN1 project. Depending on which decision is reached the tonnage bands vary greatly due to inclusion of the nanomaterial within the general dossier of the substance or within individual dossiers for each nanomaterial surface modification. In light of these legal issues pending resolution it was decided to apply all options to each case study material regardless of whether or not the production volume justifies the application under the current legal framework. The discussion of the knowledge gain and the risk minimization concerning human health and the environment within this annex is therefore a best case scenario when considered in light of risk minimization. Options that do not apply to the individual nanomaterial due to the current tonnage bands are currently not relevant in practice but might become relevant if tonnage bands for nanomaterials are shifted.

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1 Bundesanstalt für Arbeitsschutz und Arbeitsmedizin; Federal Institute for Occupational Safety and Health
2 Umweltbundesamt
3 Bundesinstitut für Risikobewertung; Federal Institute for Risk Assessment (BfR)
4 Sachverständigenrat für Umweltfragen; German Advisory Council on the Environment
2 Application of option profiles to selected case studies

The 9 options out of the 21 options proposed for the modification of REACH to enhance its applicability to nanomaterials (also referred to as nano particle where appropriate) will be analysed individually for each selected case study. The consequences of retaining the current legislative status quo and of implementing a given option will be explored and labelled as A) status quo and B) option implementation, respectively. To estimate the impact of this choice on human health and the environment, extrapolations from the current state of knowledge on the specific bulk- and nanomaterial properties as well as from the general information of nanomaterial behaviour are made. However, in many cases the knowledge on nanomaterial properties along the entire lifecycle is severely limited or lacking.

Three general considerations have direct impact on the toxicokinetics, the environmental fate and toxicity of nanomaterials. They are of particular importance beyond the actual substance the nano particles are made of and are important for every nanomaterial known up to now.³

All of them hinge on the particularly small dimension of nano particles. Knowing that 0.1 nm (1 Å) equals the size of one hydrogen atom one can imagine that a nano particle made up of a few dozen to a few hundred atoms displays these atoms in a very different fashion to the surroundings than bulk materials do. Due to the dimensions and the high curvature of the nano particle the surface atoms are highly exposed and have much fewer surrounding atoms of the same type to bind to. As a consequence the melting point of nanomaterials is severely reduced in contrast to their bulk material form (provided both have at least the same crystal structure).⁴ More importantly for the following considerations however, is the much higher surface area, made up of more exposed atoms compared to bulk material. To provide an idea of the differences between bulk and nano material surface areas: A carbon micro particle with a diameter of 60µm has a mass of 0.3 µg and a surface area of 0.01 mm². 0.3 µg of carbon in nano particle form has a surface area of 11.3 mm² if the nano particles each have a diameter of 60 nm. Considering that 0.3 µg of 60 nm sized carbon nano particles consist of 1 * 10⁹ nano particles the ratio of surface area to volume/mass for a nano particle is 1000 times higher than for a micron sized particle.⁵ This goes along with a roughly 1000 fold increase of reactivity of nanomaterials versus bulk materials.

The three specific properties are discussed thereunder:

1) The size of the primary nano particles and their agglomerates in the specific application, upon internalization, contact with the living organism, or an environmental compartment is a major determinant for the distribution, dissolution, and persistence patterns. As shown for small mammals, the body, especially the lungs, are less efficient in clearing nano particles than micrometre sized particles.⁶ In this context, interspecies variations have to be taken into account. Larger mammals are frequently more efficient at clearing nano particles from their lungs than smaller mammals (humans vs. rats).⁷ Also if nano particles gain access to the blood stream or are administrated intravenously their accumulation patterns are dependent on their size (among other determinants). Accumulation e.g. in the liver is frequently facilitated by the particle being smaller than 50 nm. In the spleen however objects...
(liposomes) of ca. 100 nm are not efficiently accumulated whereas an increase in size also led to a higher uptake rate.\(^3\)

The formation of agglomerates is another important factor which is dependent on: size and material of the original nano particle, the application the nano particle is used in, and the ubiquitous surface modifications (see p.7).

2) The protein coat that every nano particle acquires when entering a living organism\(^8\) due to its high reactivity is called corona.\(^9\)-\(^16\) Proteins binding to the nanomaterial have a profound impact on the retention time in an organism as they modulate the uptake rate in organs and thus the distribution within the organism.

The composition of the corona may vary greatly and is dependent on the properties of the nano particles, such as surface charge, morphology (curvature/size), and the substance.\(^17\) In addition, nanomaterials may associate with a specific set of proteins when subjected to different exposure routes (e.g. intravenous administration, inhalation etc.). Within the blood stream, proteins such as albumins might attach and protect the nano particles from aggregation, thus regulating retention in the blood stream, organs the nano particles accumulate in and clearance pathways. In the lung, the bronchial surfactant induces agglomeration of metal oxide nano particles and thus an increase in particle size which lowers the dispersion in the lung and enhances the clearance rate from the lung.\(^18\)

In principal, two major classes of associated proteins were shown to significantly influence toxicokinetics of nanomaterials, opsonins and dysopsonins. Opsonins are molecules that target nanomaterials for an immune response and act as a binding enhancer for phagocytosis. This leads to faster clearance from the organism by the oral route in the lung or via excretory systems in the body. Dysopsonins are proteins that lead to the opposite effect and extend the retention time in the bodily compartment the nanomaterial is currently located in. A hydrophilic, non-charged (neutral) surface disfavours the binding of opsonins to nano particles.

Additionally the small size and the extreme curvature of the nano particles are the causes for the proteins that interact with the artificial particle to be subjected to extreme interface conditions. Under many circumstances this leads to the loss of the delicate protein structure (i.e. denaturation) and thus inactivation of the protein or severe changes in protein function. The structure of the proteins, their size relative to the nano particle in question, the nano particle shape, size, material, and functionalisation (etc.) obviously all play important parts in such considerations.\(^19\)-\(^21\) In some cases also an extreme stabilization of the proteins attached to specific nano particles was reported which essentially freezes the protein in a state. Steric changes of the protein structure are however essential to the function of most proteins. ‘Freezing’ in state is just as harmful to protein function as partial or full denaturation.

In the environment humic acids can fulfil the role of opsonins and increase dispersibility.

3) The **surface charge** and hydrophobicity of a nanomaterial dictate large parts of its interactions with other molecules because its surface is so large in comparison to its volume/mass. Within blood plasma hydrophobic particles are coated with opsonins much
more quickly than hydrophilic ones.\textsuperscript{22} Positively charged particles attract different proteins than nano particles with a negative surface charge. This is due to the protein specific isoelectric points. In general hydrophobicity and surface charge are key determinants for the selection of surface bound proteins. To a lesser degree the substance of the nano particles, the shape and the size also influence the selection process. The coating of the nano particles heavily influence aggregation, motility, bioavailability and retention time within any compartment of the living organism that the nano particles are located in.

These three general considerations are applicable to all nanomaterials and, as hinted at above, they are the reason why \textit{nano particles share certain non-substance specific, toxicological characteristics} when living systems are exposed to them. These toxicological properties are, broadly put, size specific. If the nano particles are soluble then these basic considerations apply for the lifetime of the nano particle form and are changed and then lost over time together with the physical shape. Because solubility is heavily dependent on the environment and the substance of the nano particle total dissolution of the nano particle might happen quickly thus rendering size specific effects at the dissolution location unimportant. The dispersion to a location where the conditions are favourable for dissolution is still dependent on the parameters imposed by the small size of the nano particles. If the dissolution of the nano particle proceeds very slowly then the size specific effects described in this chapter are in effect for a longer time and become relevant for risk assessments.

The hardest property to quantify in hazard assessments is the ability of nano particles to act as vectors or shuttles\textsuperscript{23} for a broad variety of chemicals including bacterial toxins. In conjunction with toxic chemicals the toxic effects can be enhanced due to better access to the organism when bound to nano particles much as the targeted deliverance of pharmaceuticals to sites of pathology inside an organism can be optimized with the aid of nano particles.\textsuperscript{24} The variety of potential interaction partners of ‘luggage’ for a given nano particle depends heavily on its lifecycle.

nano particles inhaled by air and/or water breathing organisms are prone to create similar effects in connection with the sensitive membranes that compromise breathing apparatuses in large parts of the animal kingdom. Excessive exposure to nano particle-matter can cause lung overload and/or inflammation due to inflammatory reactions via secondary generation of reactive oxygen species (i.e. oxidative stress).\textsuperscript{25–27} nano particles are also able to trigger fibrotic responses and carcinogenic effects if inhaled chronically at large enough doses. The size distribution of inhalable nano particles has serious impacts on their inflammatory potency and their persistence time in the respiratory tract\textsuperscript{28}, because it affects the distribution in the breathing apparatus. Smaller nano particles with less agglomeration tendency penetrate much deeper into the organ than larger or strongly agglomerated particles. The individual effects obviously vary in their severity of disease symptoms from cell type to cell type\textsuperscript{29}, species to species, in a dose-, and exposure dependent manner.

Beyond these general effects of nano particles, individual nano particle substances may have additional, substance specific toxic effects of varying severity.\textsuperscript{30} These will be discussed below in the context of the selected case studies.
Also the ligands that are accumulated by the nano particles on their surfaces during the course of their lifecycle will vary from substance to substance and pose specific challenges with regard to the hazard assessment for each nano particle substance (see below). The exposure scenarios can significantly from substance to substance and from application to application.

The high surface area of nano particles makes them particularly susceptible to surface modifications (the corona described above is a special, protein related case). In fact every nano particle is surface modified. This is true from the first contact of atmosphere and nano particle throughout the life cycle of the product, because of the high surface area and consequently the high reactivity of the nano particles and their agglomerates. In the discourse on safety of nanomaterials the awareness of intentional vs. unintentional surface modifications is essential. The impacts of intentional surface modifications are substantial and often well characterized. They are used to maintain granular distributions, mediate targeting in medical applications, convey photochemical properties etc. However, because of the high reactivity of nano particles described above, pre-imparted surface treatments and modifications are easily altered, lost or substituted along the life cycle of any nano particle. Current studies on nano particles suggest taking into account, that intermediate or final forms of the nano particle are neither in the same agglomeration / aggregation state, nor are they likely to carry the same unintended surface modifications. Such alterations of nano particle surface chemistry have profound impacts on toxicological and exposure related considerations.

Modifications that provide hydrophilicity or hydrophobicity are among the most straight forward impacts on the nano particle distribution in the organism and the ecosystems. The shades of gray between these two extremes and the additional properties that might be imparted by different ligands such as membrane permittivity, bioreceptor recognition, the accumulation of organochemical toxins on the nano particle surface, loss of solubility for specific solvents, as well as the potential for the nano particles to become available for the accumulation in the food chain are examples for some of the many possible modifications with harmful potentials towards human population and ecosystems.

A thorough understanding of the physico-chemical and toxicological properties of nano particles with their intentional surface modifications is the important first step to encompassing and controlling the inherent hazard. The second step consists of understanding which unintentional modifications are imparted and the properties they convey to the nano particle during its application life cycle.

It is important to stress that the very same modifications might also prove to be desirable as they might assist in removing the nano particles from major exposure routes by significantly changing the agglomeration state or inducing the formation of complexes that are less toxic. Full life cycle analysis (LCA) would however be the only way to conclusively show positive or negative effects.

The 21 options proposed for the modification of the current REACH regulation provide a basis towards ensuring a responsible and complete risk assessment. The following section outlines the potential impacts on human health and the environment, as exemplified by detailed case studies on nano particles of: zinc oxide (ZnO nano particles), titanium dioxide (TiO₂ nano particles), and nano diamond). Seven additional examples are presented in brief.
3 Methodology for the qualitative assessment of the exposure and risk associated to selected nano particles

We decided to follow accepted evaluation patterns by dividing the hazard assessments for the individual nanomaterials into human and environmental hazards. Human health hazards are grouped into acute effects, chronic effects and mortality. Environmental hazards were grouped similarly into acute and chronic effects but as these two groupings frequently include studies on mortality we added a third category covering accumulation/persistency to address long term effects of the specific nanomaterial. In this regard accumulation and persistency are of particular concern as the food chain magnifies the toxicological concerns in dependence of those two aspects.

The considerations above are briefly illustrated in the flow charts below and the considerations elaborated in the risk and impact assessments below follow these patterns.

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Figure 1: Grouping of risk and exposure data for humans and the environment
4 Detailed case studies on the impacts of the individual options on selected nanomaterials

4.1 Titanium dioxide nano particles

As with all nanoparticles there are certain general toxicological characteristics that TiO$_2$ nano particles (*we address specifically TiO$_2$ nano particles and will use this short form henceforth*) share with other nano particles. These have been extensively elucidated in the introduction.

Beyond these general hazards inherent to inhalable nanomaterials TiO$_2$ nano particles have an additional property to consider. In contrast to ZnO nano particles TiO$_2$ nano particles can be manufactured in different crystal forms. Rutile and anatase are mainly manufactured where rutile is the preferred crystal form in products with consumer exposure as it generates less ROS than anatase. Anatase is however the commercially most used isofrom. $^{37}$ Both however are able to generate ROS directly with the aid of UV and visible light. $^{38}$ Especially with unicellular, light dependant organisms such as microalgae or soil bacteria this property magnifies the hazard posed by TiO$_2$ nano particles.

**Applications**

TiO$_2$ nano particles are used in a large amount of products covering all states from tight integration into complex materials to nearly free (agglomerated) nano particles. It was produced at tonnage bands of more than 50,000 t/y in 2010. By 2015 the production is projected to increase to 201,500 t.$^{39}$

TiO$_2$ nano particles find their applications in products where they provide UV-protection, UV-absorption for catalytic processes, or serve as pigments in paints and varnishes. The ability to absorb UV light and redirect the collected energy is used in sunscreens (main consumer exposure). $^{40}$ Here the collected energy is redirected towards forming other substances than ROS if possible.$^{41,42}$

The high opacity of the white created by TiO$_2$ particles is used in paints and varnishes among other applications. Here TiO$_2$ nano particles also serve as one of the main constituents for the ‘self-cleaning’ properties newer coatings and materials have. In that case the ROS production is desirable in order to destroy adherent dirt. It should be noted however that applications listing TiO$_2$ as a pigment are estimated to contain less than 0,1 % (w/w) TiO$_2$-nano particles by the IARC.$^{43,44}$ The same function is used in antibiotic coatings of textile fibres and to cleanse water in waste water applications.

**Physico-chemical properties**

TiO$_2$ nano particles occur in three crystalline structures: Anatase (tetragonal crystals), brookit (orthorhombic crystals, not manufactured commercially) and rutile (tetragonal crystals). As mentioned above TiO$_2$TiO$_2$O$_2$ is able to absorb UV light. In nano particle form TiO$_2$TiO$_2$ is transparent in the visible spectrum which makes it desirable for applications such as sunscreen lotions. In these applications the rutile form is preferred. The anatase form of TiO$_2$ nano particles has specific electric, photocatalytic and thus also antimicrobial properties. Coupled to the capacity to form ROS when
exposed to UV and visible light is also the ability to form other – potentially undesirable – side products such as formaldehyde, acetic aldehyde and nitrates. These pose additional risks for human health and the environment.

Exposure

The exposure of humans and the environment to TiO₂ nano particles can occur at all stages of the TiO₂ nano particle life cycle. The likelihood of exposure during the production is comparatively low, if work places are equipped with appropriate measures. In 2011 Kuhlbusch et al. were unable to detect significant exposure at workplaces processing TiO₂ nano particles.⁴⁵ Hansen et al. estimate a daily exposure of 57 µg/kg*day for a two year old from TiO₂ nano particles contained in sunscreen lotions (dermal exposure).⁴⁶ The TiO₂ nano particles are not able to transverse the barrier the skin poses. Even inflamed or otherwise damaged skin poses an effective barrier to these nano particles.

Some sunscreen products are also offered as a spray application. Inhalative exposure to TiO₂ nano particles and larger particles was calculated to be 3.5 g/m³ in 2011.⁴⁷ Additionally Powell et al. (2010)⁴⁸ estimate a daily oral intake of micro and nanosized titanium in Great Britain at around 5 mg/person due to additives in food added for colouring effects (here nanosized refers to a size range of 100-200 nm).

The main sources of TiO₂ nano particles in the environment are currently judged to be the use of sunscreens, and the washing-out of nano particles from house paint and the wear imposed on coated surfaces of construction materials and coated windows. A significant percentage of the nano particles in house paint are lost in the course of 2 years merely due to environmental effects.⁴⁹ Surface waters in Europe were estimated to carry between 0.7 µg/L and 24.5 µg/L whereas the exposure of soil and sediments was estimated to be between 1 and 1.03 g/kg.⁵⁰,⁵¹ TiO₂ in any size range is not water soluble and tends to agglomerate in water. This does not significantly affect its reactivity but the distribution in the environmental compartments might be affected because of the sedimentation processes, that gain impact with increasing particle size. Once the TiO₂ nano particles have sedimented it is unlikely that they become mobile again.⁵² Kiser et al. (2009) have shown that part of the TiO₂ nano particles brought into a wastewater treatment plant are not held back in the sludge but are released to the environment.⁵³

Toxicology

Beyond the general dangers (acute as well as chronic) listed above that inhalable nano particles potentially pose TiO₂ nano particles are suspected to interact with the DNA and thus have direct genotoxic potential as they have been found inside cellular nuclei in lungs and in cultured cells.⁵,⁵⁴–⁵⁶ These results are interesting, but it is important to note that the studies yielding these results have been carried out with high concentrations of TiO₂ nano particles. Based on inhalation studies in rats TiO₂ nano particles are rated as potentially carcinogenic (group 2B) by the IARC.⁴³,⁴⁴ Dermal absorption of TiO₂ nano particles could not be shown experimentally, thus making toxicological...
effects due to sunscreens etc. unlikely. Oral intake of TiO₂ nano particles results in exposure of the gastro intestinal tract and may result in accumulation in internal organs. In the presence of UV light TiO₂ nano particles are significantly more toxic due to their photochemical reactivity.

**Ecotoxicology**

There is little reliable data available concerning the effect of TiO₂ nano particles on terrestrial organisms. Studies have been made with a focus on the effects on aquatic organisms. The lack of clear characterization of the test material and consistent experimental conditions however make it difficult to draw any conclusions as the results are contradictory. The German Federal Environment Agency and the Danish Ministry of the Environment both published studies correlating data of different experiments on the model organism *Daphnia magna*, the results of which range from nontoxic to toxic for TiO₂ nano particles.

Experiments with rainbow trout show little acute toxicity but also reveal morphological changes in gills, the intestinal tract. Oxidative stress was detected in these organs as well as in the brain (see also studies cited above). Furthermore there is experimental evidence of Arsenic and Cadmium accumulation in context with TiO₂ nano particle presence in fish. This might be due to the association of the heavy metals with the nano particles thereby transforming the nano particles into effective carriers. There are studies that indicate bioaccumulation processes for TiO₂ nano particles in aquatic organisms but further tests are necessary to verify the existing data.

Crustaceans have been exposed to TiO₂ nano particles using the sample organism *Daphnia magna* and acute as well as chronic effects were detected. Due to the widespread results it is difficult to exactly determine the toxicological profile in crustaceans.

Single cellular organisms such as bacteria, certain algae and yeast have also been exposed to bulk TiO₂ and TiO₂ nano particles. The toxicity varies considerably and is also enhanced in the presence of UV light due to the additional production of ROS. Gram-positive bacteria (*B. subtilis*) are more susceptible to the antimicrobial activity of TiO₂ nano particles than gram-negative bacteria (*E. coli*).

Aruoja et al. (2008) showed that the microalgae *Pseudokirchneriella subcapitata* is significantly more susceptible for toxic effects originating from TiO₂ nano particles (EC50=5.83 mg/l) than to effects from bulk TiO₂ (EC50=35.9 mg/l). To yeast neither bulk TiO₂ nor TiO₂ nano particles were toxic even at concentrations of up to 2,000 mg/l. These results show that the effects of nano particles can vary strongly from one species to another and very specific requirements and questions have to be applied to each nanomaterial.

To our knowledge three studies have been published thus far on bioaccumulation of TiO₂ nano particles. In *Daphnia magna* relatively slow elimination of TiO₂ nano particles with an average size of 21 nm diameter led to accumulation. Rainbow trout were exposed to TiO₂ nano particles by Ramden et al. (2010) and they found TiO₂ accumulation in gut, liver, spleen, gills, and brain after 8 weeks of exposure. Scown et al. (2010) found bioaccumulation of TiO₂ in the kidneys where the nano particles remained for up to 21 days after intravenous exposure. Clearance was noted after 90 days from the kidneys but the TiO₂ nano particle concentrations found in the liver did not change. Retention of TiO₂ nano particles was longer than the runtime of the experiment.
Grading in the hazard table (see Final Report)

TiO$_2$ nano particles are graded with a medium hazard albeit being listed as potentially carcinogenic (group 2B) by the IARC as they consistently exhibit the toxicity that can be attributed to all inhalable nanomaterials. Their ability to generate ROS varies in its intensity with the crystal structure (anatase, brookit, rutile) but is highly dependent on the availability of visible or UV light. Environmental experiments yield contradictory results with regard to the toxicity in crustaceans. They show a tendency for the TiO$_2$ nano particles to bio-accumulate, but the acute toxicity is again coupled to the availability of visible or UV light which is only given in single cellular organisms living in exposed habitats. The accumulation in higher organisms led to oxidative stress and morphological changes being detected in rainbow trout for example. TiO$_2$ nano particles lack the ionic effects that pose additional danger from other nanomaterials.

4.2 Impact of the options on risk and exposure scenarios associated to titanium dioxide nano particles

The following tables illustrate where option implementation or lack thereof will affect the fields listed below without implying positive or negative nature of the impact. The tables serve as an orientation for the reader to illustrate those aspects that will be of particular interest in the following discussion and the ensuing upscaling of the impacts of the proposed modifications to REACH.

Table 4-1: Human health impacts of TiO$_2$ nano particles

<table>
<thead>
<tr>
<th></th>
<th>options with impacts</th>
<th>pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>acute effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,11,12,21</td>
<td>local inflammation of the lung</td>
</tr>
<tr>
<td>consumers</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td>via the environment</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td><strong>chronic effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,12, 21</td>
<td>fibrotic changes in the lung tissue, systemic accumulation in organs such as kidney, liver, brain, etc.</td>
</tr>
<tr>
<td>consumers</td>
<td></td>
<td>accumulation of TiO$_2$ nano particles via the food chain</td>
</tr>
<tr>
<td>via the environment</td>
<td>6,13,16,17,18,19,21</td>
<td></td>
</tr>
<tr>
<td><strong>mortality</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,12,21</td>
<td>cancer</td>
</tr>
<tr>
<td>via the environment</td>
<td>6,13,16,17,18,19,21</td>
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Table 4-2: Environmental impacts of TiO$_2$ nanoparticles

<table>
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<th>options with impacts</th>
<th>effects</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Acute effects</strong></td>
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<td></td>
</tr>
<tr>
<td>soil and sediment</td>
<td>6,13,18,21</td>
<td>bactericide</td>
</tr>
<tr>
<td>organisms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>aquatic organisms</td>
<td>6,13,17,21</td>
<td>bactericide, algaecide, pathogenic to higher</td>
</tr>
<tr>
<td></td>
<td></td>
<td>organisms (fish, crustaceans), increased</td>
</tr>
<tr>
<td></td>
<td></td>
<td>mortality rates</td>
</tr>
<tr>
<td><strong>Chronic effects</strong></td>
<td></td>
<td></td>
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<tr>
<td>soil and sediment</td>
<td>6,13,18,21</td>
<td>bactericide</td>
</tr>
<tr>
<td>organisms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>aquatic organisms</td>
<td>6,13,17,21</td>
<td>pathogenic to higher organisms (fish,</td>
</tr>
<tr>
<td></td>
<td></td>
<td>crustaceans), increased mortality rates</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>**Accumulation/</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Persistency**</td>
<td></td>
<td></td>
</tr>
<tr>
<td>soil and sediment</td>
<td>6,13,21</td>
<td>persistent</td>
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<tr>
<td>aquatic sphere</td>
<td>13,21</td>
<td></td>
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<tr>
<td>biota</td>
<td>6,13,17,18,21</td>
<td>bioaccumulation (see effects above)</td>
</tr>
</tbody>
</table>

4.3 Impact of the option implementation to REACH on risk and exposure (TiO$_2$ nanoparticles)

Option 6 – Include information on dustiness

A) Status quo
The need for inhalation studies in the endpoint section is not apparent which has the potential to lead to incomplete exposure assessments. The implications for workers, consumers and ecological habitats are considerable as damage has to become apparent before further research is initialized. Furthermore this option specifies the need for these studies independent from the production or import quantities. Inhalation studies are otherwise required at a production/import tonnage of 10 t or more.

Without the data gained from inhalation study the need for specialized air filters and breathing masks at exposed workplaces might not become apparent.

B) Option implementation
TiO$_2$ nanoparticles are a dusty substance and risk assessments as well as necessary endpoint related studies are determined by this property. Inhalation studies with various size distributions and in
different environments are necessary and the obtained data will be applied in security considerations along the lifecycle of products containing TiO$_2$ nano particles in inhalable form.

**Option 11 – Require acute toxicity data for the most relevant route of exposure.**

The toxicity data provided by the registrant should be coupled to the most relevant route of exposure of humans to a given substance and nanoform.

The amount of TiO$_2$ nano particles produced requires the full range of toxicity tests and inhalation is clearly a relevant exposure route. This option has no impact on TiO$_2$ nano particles.

**Option 12 – Change particles to ‘(nano)particles’ for repeated dose toxicity studies (inhalation)**

This option relates to the phrasing in the Annexes VIII, 8.6.1, IX, 8.6.2, and IUCLID section 8.5.2. The boundaries of the definitions here should be extended and specified by exchanging all instances of the word ‘particles’ for ‘(nano)particles’. Furthermore the following addition should be considered:

‘Especially for (nano)particles it should be justified when inhalation is not considered a relevant route of exposure.’

Either the test methods (for sub-acute, sub-chronic or combined repeated dose toxicity/reproductive screening study) or the guidance should refer to extended pathology/histology determinations and examination of relevant parameters in BAL (bronchoalveolar lavage) fluid.

**A) status quo**

Without the imposed fine resolution of the inhalation studies with regard to the employed size ranges of the nano particles and their modifications, little information is gained beyond the particular experimental setup. Specific applications and the alterations to the surface properties of the nano particles during the usage are the first ‘experiments’ indicating hazards to humans

**B) option implementation**

As stated for the option 11 here also the option has little impact on the practice as TiO$_2$ nano particles are already tested for inhalation toxicity due to the volume of production. In relation with previous options however a finer detail is achieved here by testing explicitly the nanoforms of TiO$_2$ for health risks connected to inhalation in dependence from their size distribution. Hazards connected to TiO$_2$ nano particle applications in which inhalation is imaginable are well characterized thus further minimizing the risk to humans. Especially work place related hazards are detected early.

**Option 13 – Require non-bacterial, in vitro gene mutation study**

Genotoxicity of TiO$_2$ nano particles has been shown in various studies (see introduction TiO$_2$ nano particles) although primary-, secondary-ROS generation, and direct particle DNA interaction have so far not been sufficiently distinguished to determine the cause of genotoxicity.
**Option 16 – Consider water solubility in relation to test waving**

Nano particles are often not soluble in water. The lack of water solubility is not a reliable indicator for the lack of bioavailability. Nano particles have other options available to them to interact with their environment and biological cells.

**A) status quo**

TiO\(_2\) nanoparticles are not water soluble and thus testing of bioavailability, -accumulation, and persistence can be waived. In all three aspects solid data shows TiO\(_2\) nano particle activity/presence in spite of the lack of solubility per se.

**B) option implementation**

TiO\(_2\) nanoparticles are tested for all relevant endpoints and accurately described for hazard and risk assessment within humans, other organisms, and the environment. As numerous results show the lack of water solubility does not correlate with unavailability in the biosphere. This is especially true for nano particles.

**Option 17 – Specify that long term testing should not be waived based on lack of short term toxicity.**

Some nanoforms have a tendency to accumulate in biological systems and should thus not be discounted when considering toxicity just because they do not exhibit short term toxicity. TiO\(_2\) nanoparticles exhibit short and long term toxicity in all organisms exposed to them with the exception of yeast which one study shows to be impervious. This option has no effect on TiO\(_2\) nano particles.

**Option 18 – Require that testing on soil and sediment organisms is prioritised**

**A) status quo**

As TiO\(_2\) nanoparticles are not soluble the deposition in soil and sediments is relevant but potentially overlooked. Bioaccumulation in the soil and in sediments might play an important role in creating a hazardous situation over time.

**B) option implementation**

The exposure characteristics of TiO\(_2\) nano particles are easily altered by changing surface modifications in the course of the TiO\(_2\) nano particle lifecycle. The testing of soil and sediment organisms will quantify the amount of hazard in relation to surface modifications and in dependence of applications of the TiO\(_2\) nano particles. These data are interlinked with the assessment of bioaccumulation processes in this habitat.

**Option 19 – Specify that algae testing should not be waived based on insolubility**

See Option 16
Option 21 – Require considerations of most appropriate/relevant metrics

The appropriate metrics should be chosen also to develop and present a realistic exposure assessment and risk characterization. Mass metrics are not necessarily appropriate as a point of reference for the evaluation of nanomaterials.

4.4 Zinc Oxide Nano particles

The specific properties of ZnO nano particles will be briefly highlighted below. As described above, some toxicological properties are specific to the substance the nano particle is made of and are cumulative with the general considerations described above. The impact of the individual options for the modification of REACH will become more apparent with the information given here in mind.

Applications

33.400 t/y of ZnO nano particles were produced worldwide in 2011. It is used in micro and nanoform as a UV protection agent. As such, it is a functional part of sunscreen lotions, cosmetic products, paints and varnishes. It is used in plastics and rubber where it is embedded in the product matrix and finds applications in scientific research and the medical sector as bactericide and in the treatment of cancer. Furthermore, ZnO nano particles are used in the electronics industry for the production of e.g. semiconductors or flexible TFT-screens.

Physico-chemical properties

ZnO in every form dissociates in aqueous environments releasing Zn$^{2+}$ ions. Zincoxide is able to absorb UV-A and UV-B irradiation. The harvested energy can be employed to produce reactive oxygen species (ROS). ZnO nano particles in comparison to coarser particles do not scatter light in the visible spectrum and thus appear translucent.

Exposure

The main exposure routes for to humans and the environment are determined by the extensive usage of ZnO nano particles as UV protection agent. Humans can be exposed to ZnO nano particles either via direct, topical application on the skin in the form of cosmetics and sunscreens or unintentionally via the use of paints and varnishes (workers and amateurs alike). The latter are likely to release nano particles to the environment as was also shown for nano silver and titanium dioxide nano particles. It is also possible that ZnO from coatings is lost via ion leaching. Further research is necessary to determine the exposure arising from such applications. Open applications such as cosmetics easily release ZnO nano particles with or without specific surface modifications to the environment. In all likelihood, nano particles from the applications mentioned above end up in agglomerated or actual nano particle form in the wastewater system from where a fraction is distributed into the ecosystem.
The complexed forms of the ZnO nano particles found in e.g. plastics and rubber pose no particular risk to consumer health as they are usually not released from their matrix and consequently are not bioavailable.

At the workplace the classical exposure route is possible during welding processes. However, in modern industrial workplaces protective measures are set in place. In Germany a maximum workplace concentration of airborne ZnO nano particles is set at 1 mg/m³.\(^77\)

Model calculations of ZnO nano particles in the surface water bodies of Europe span a wide range between 0.03 µg/l and 76 µg/l.\(^51\)

**Toxicology**

The ion release into the aqueous surroundings (including tissues and cells) is the main toxicological effect to be considered in humans and the environment.\(^64,65,70,78\) In general, ion leaching is dependent on the particle stability which is heavily influenced by the coating of the nano particles among other factors. The ZnO nano particles themselves are able to generate ROS much like other nano particles.\(^79\)

Beyond the basic set of toxicological concerns common to all inhalable nano particles (see introduction), ZnO nano particles are not perceived as particular risk to humans yet. This is due to the fact that Zinc is an essential trace element. A daily dose of around 70 mg/day and individual human does not lead to any adverse health effects. Up to this dose and with the oral uptake route the ions do not represent a problem for humans.\(^36\)

However, if humans are exposed to airborne ZnO nano particles in relevant doses they are affected by metal fever. The symptoms appear as influenza like pathology resulting from the inflammation of the lung that is reversible within 48 h if no further particles are inhaled. Consumers are rarely exposed to fumes or inhalable dust containing ZnO nano particles. Only workers in the particular field of industry or researchers have an increased chance of being exposed to large enough doses of inhalable dust. To our knowledge, the first scientific documentation of this pathology was presented in 1927.\(^80,81\) Furthermore, the chronic inhalation or the overload of the lung with dusty nano particles can lead to fibrotic changes in the lung and consequently to cancer (most chronic effects are likely due to be particle related effects with added impact due to nano particle distribution patterns in living systems). Due to the high dissolution rate of ZnO nano particles sufficient concentrations for particle effects to present themselves would only be achieved via coatings that decrease the rate, via chronic exposure, or via a large initial dose. In case of a large dose the dissolution equilibrium would have to be reached before the nano particles dissolve entirely. As Xia et al. (2008) have shown ZnO nano particles triggered all three tiers of oxidative stress and are thus significantly more effective in this regard than TiO\(_2\) nano particles.\(^28,36,70,82,83\)

In the case of uncoated (= pristine; *relative term – see discussion on unintentional surface modifications p. 7*) ZnO nano particles, current research attributes the pathologic effects to two sources. Firstly, the nano particle-form of zinc makes it inhalable for humans when the nano particles are airborne.\(^84,85\) Secondly, ZnO nano particles release Zn\(^2+\) ions in aqueous environments. These conditions are found inside living organisms and cells as well as in aquatic habitats. The dissolution processes are also responsible for the toxicity rating of bulk ZnO\(^78,86\) and are responsible for altered
exposure patterns. The high surface area of ZnO nano particles can benefit the release of Zn$^{2+}$ ions in comparison to bulk sized ZnO. If ZnO nano particles are inhaled or are absorbed in the gastrointestinal tract the effects of the leached ions become more pronounced. Zn$^{2+}$ ions are able to interact with cellular structures and proteins by either inactivating or denaturating them. This is a stochastic process with many potential cellular targets. The same is true for the generation of ROS and the effects of the radicals on the surrounding biomolecules.

The acquisition of a corona upon ingestion or inhalation may profoundly change the distribution patterns of ZnO nano particles in the human body, further complicating the assessment of associated risks. There is some evidence, however, that inhalation and interaction with the BAL (bronchoalveolar lavage; lung fluids) leads to nano particle agglomeration and thus clearance from the lung is facilitated.

The most prominent exposure of consumers to ZnO nano particles is via their skin through the application of cosmetics and sunscreens. No negative effects have been documented to the present time. ZnO ions and nano particles are able to penetrate the outer skin barrier to a limited degree that is not considered to be a cause for concern.

**Ecotoxicology**

Because of the increasing industrial and cosmetic applications ZnO nano particles are used in, it is likely that their concentration in the environment will rise in the future. Zn$^{2+}$ ions are the main concern in the environment alongside with the potential of Zn$^{2+}$ ions to accumulate in the food chain. Zinc (ZnO) in bulk form is listed as hazardous substance, with particular danger to aquatic organisms and the potential to inflict long term damage to aquatic habitats (Category 1 H400, H410 GHS; WGK 2, Germany; CLP Index No.: 030-013-00-75). ZnO nano particles tend to agglomerate in the aquatic compartment. This effect is further enhanced by the presence of organic interaction partners. Within this compartment they are able to release Zn$^{2+}$ ions to the surrounding water. The ZnO nano particles are persistent in soil and sediments. Ion release is higher from nano particles than from bulk material due to the increased surface area of the ZnO nano particles. Additionally the distribution of ZnO nano particles is altered compared to the distribution of bulk ZnO because of the higher dispersion rate inherent to nanomaterials. The potential for the acquisition of unintentional surface modifications of the ZnO nano particles is hard to determine into hazard considerations because there is little research along the life cycle of ZnO nano particle containing products available at this point. Bacteria and higher organisms such as crustaceans and fish can be heavily affected by rising Zn$^{2+}$ ion concentrations resulting in increased adult and embryonal mortality and embryonal deformations. The NOEC for crustaceans was set to 30 µg/l. In soil and sediments ZnO nano particles are persistent for up to 14 days and here also displayed their negative effects on the reproduction rate of worms and the survival of soil bacteria.

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5 Classification, Labelling and Packaging; http://clp-inventory.echa.europa.eu/SummaryOfClassAndLabelling.aspx? SubstanceID=93&HarmOnly=no?fc=true&lang=en
Grading in the hazard table (see Final Report)

ZN nano particles were graded as highly hazardous because even without the unintended, potentially harmful surface modifications that the nano particles can pick up during their life cycle, ZN nano particles already exhibit high toxicity mainly via the effects of their ions in the environment. Especially in the aquatic environment their nano-morphology just heightens the ion related effects. In addition zinc oxide acquires pathological properties upon inhalation, ingestion and the potential to disperse even wider systemically as well as environmentally when in nano-morphology. Counting in unintended modifications that impart an ubiquitous hazard to all nano particles some known modifications can lead to retardation in the ion leaching process and higher dispersibility in the aquatic habitat or to increased retention times within organisms for example.

4.5 Impact of the options on risk and exposure scenarios associated to zinc oxide nano particles

The following tables illustrate where the implementation of the additional options or lack thereof will affect the fields listed below without implying a positive or negative nature of the impact. The tables serve as an orientation for the reader to illustrate those aspects that will be of particular interest in the following discussion and the ensuing upscaling of the impacts of the proposed modifications to REACH.

Table 4-3: Human health impacts of zinc oxide nano particles

<table>
<thead>
<tr>
<th></th>
<th>options with impacts</th>
<th>pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>acute effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,11,12,21</td>
<td>influenza like symptoms; 48 h, local inflammation of the lung</td>
</tr>
<tr>
<td>consumers</td>
<td>21</td>
<td>see above</td>
</tr>
<tr>
<td>via the environment</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td><strong>chronic effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,12,13,21</td>
<td>fibrotic changes in the lung tissue</td>
</tr>
<tr>
<td>consumers</td>
<td>6,21</td>
<td>accumulation of ZnO nano particles via the food chain</td>
</tr>
<tr>
<td>via the environment</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td><strong>mortality</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,12,13,21</td>
<td>fibrotic changes in the lung tissue, cancer, unknown effects of distribution and accumulation in the body due to surface modifications</td>
</tr>
</tbody>
</table>
Table 4-4: Environmental impacts of zinc oxide nano particles

<table>
<thead>
<tr>
<th></th>
<th>options with impacts</th>
<th>effects</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>acute effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>soil and sediment</td>
<td>6,13,18,21</td>
<td>bactericide, increase in mortality rates for key indicator organisms</td>
</tr>
<tr>
<td>organisms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>aquatic organisms</td>
<td>6,13,17,18,21</td>
<td>bactericide, algicide, toxic to higher organisms (fish, crustaceans), increased mortality rates</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>chronic effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>soil and sediment</td>
<td>6,13,21</td>
<td>lowering of animal reproductive rates, bactericide</td>
</tr>
<tr>
<td>organisms</td>
<td></td>
<td></td>
</tr>
<tr>
<td>aquatic organisms</td>
<td>6,13,17,18,21</td>
<td>lowering of reproductive rates, deformations</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>**accumulation/</td>
<td></td>
<td></td>
</tr>
<tr>
<td>persistency**</td>
<td></td>
<td></td>
</tr>
<tr>
<td>soil and sediment</td>
<td>6,21</td>
<td>persistent</td>
</tr>
<tr>
<td>biota</td>
<td>6,17,18,21</td>
<td>bioaccumulation (see effects above)</td>
</tr>
<tr>
<td>aquatic sphere</td>
<td>6,21</td>
<td>increase of ZnO\textsubscript{2}+ ion concentration</td>
</tr>
</tbody>
</table>
4.6  Impact of the option implementation to REACH on risk and exposure (ZnO nanoparticles)

Option 6 – Include information on dustiness (Annex VI)

A) status quo
The need for inhalation studies in the endpoint section is not apparent. The implications for workers, consumers and ecological habitats are considerable as damage has to become apparent before further research is initialized. Furthermore this option specifies the need for these studies independent from the production or import quantities. Inhalation studies are otherwise required at a production/import tonnage of 10 t or more.
Without the data gained from inhalation study the potential need for air filters and breathing masks at exposed workplaces might not become apparent.

B) option implementation
ZnO nanoparticles is a dusty substance and risk assessments as well as necessary endpoint related studies are determined by this property. Inhalation studies with various size distributions and in different environments are necessary and the obtained data will be applied in security considerations along the lifecycle of products containing ZnO nanoparticles in inhalable form.

Option 11 – Require acute toxicity data for the most relevant route of exposure (Annex VII)
The toxicity data provided by the registrant should be coupled to the most relevant route of exposure of humans to a given substance and nanoform.
The amount of ZnO nanoparticles produced requires the full range of toxicity tests and inhalation is clearly a relevant exposure route. This option has no impact on ZnO nanoparticles.

Option 12 – Change particles to ‘(nano)particles’ for repeated dose toxicity studies (inhalation)
This option relates to the phrasing in the Annexes VIII, 8.6.1, IX, 8.6.2, and IUCLID section 8.5.2. The boundaries of the definitions here should be extended and specified by exchanging all instances of the word ‘particles’ for ‘(nano)particles’. Furthermore the following addition should be considered:
‘Especially for (nano)particles it should be justified when inhalation is not considered a relevant route of exposure.’
Either the test methods (for sub-acute, sub-chronic or combined repeated dose toxicity/reproductive screening study) or the guidance should refer to extended pathology/histology determinations and examination of relevant parameters in BAL (bronchoalveolar lavage) fluid.
A) status quo
Without the imposed fine resolution of the inhalation studies with regard to the employed size ranges of the nanoparticles and their modifications, little information is gained beyond the particular experimental setup. Specific applications and the alterations to the surface properties of the nanoparticles during the usage are the first ‘experiments’ indicating hazards to humans.

B) option implementation
As stated for the option 11 here also the option has little impact on the practice as ZnO in powder form is already tested for inhalation toxicity. In relation with previous options however a finer detail is achieved here by testing explicitly the nanoforms of ZnO for health risks connected to inhalation in dependence from their size distribution. Hazards connected to ZnO nano particle applications in which inhalation is imaginable are well characterized thus further minimizing the risk to humans. Especially work place related hazards are detected early.

Option 13 – Require non-bacterial, in vitro gene mutation study (Annex VII)
Bacterial mutagenicity tests are widely used to assess basic mutagenic properties of a given compound but are not always appropriate to do so for nanomaterials. As elucidated in RIP-oN2 and ECHA-12-G-03-EN bacterial tests might lead to false negatives because inorganic (nano)particles cannot permeate the bacterial wall and membrane, thereby creating false results. Instead an in vitro test for the mutagenicity of nanomaterials is suggested.

A) status quo
If genotoxicity is dependent on the size distribution of the ZnO nano particles genotoxicity would only show up under very specific circumstances which are dissimilar for bacteria and eukaryotes. Tests with living organisms are also often confounded by the fact that the secondary ROS production triggered by the inflammatory effects of nano particles in general can lead to gene deregulation. This is then however due to the presence of the nano particle as an irritant and not a danger that is related in particular to ZnO nano particles. Without non-bacterial tests, it is impossible to assess potential genotoxicity inherent to the ZnO nano particles in particular.

B) option implementation
ZnO nano particles are not known to be genotoxic up to this point. It is unclear however if this is entirely true until non-bacterial tests are documented. The results dictate the appropriate risk prevention measures.

Option 16 – Consider water solubility in relation to test waving
Dissolution of ZnO-Ions is well known also for bulk ZnO and constitutes one of the two identified routes of toxicity. The consequences of this option do not apply to ZnO nano particles.
Option 17 – Specify that long term testing should not be waived based on lack of short term toxicity.

Some nanoforms have a tendency to accumulate in biological systems and should thus not be discounted when considering toxicity just because they do not exhibit short term toxicity. ZnO nano particles and bulk-ZnO both exhibit aquatic short and long term toxicity. This option has no effect on ZnO nano particles.

Option 18 - Specify that algae testing should not be waived based on insolubility

As described above the lack of water solubility should not be a valid reason for the waiving of aquatic tests in the case of nanomaterials. Many are able to persist in aqueous media and influence their surroundings via agglomeration or surface modification with molecules found in the environment. This issue is also taken up by the ECHA in their new guidance document ECHA-12-G-05-EN.

This option has no effect on the ZnO nano particle testing.

Option 19 – Require that testing on soil and sediment organisms is prioritised

A) status quo

The exposure characteristics of ZnO nano particles are easily influenced by changing surface modifications in the course of the ZnO nano particle lifecycle. While the nano particles will also end up in soil and sediments (if their modifications slow down the dissolution) the main toxicity arises from the dissolution in aqueous environments. The testing of soil and sediment organisms will quantify the amount of hazard in relation to surface modifications and in dependence of applications of the ZnO nano particles. The impact on soil and sediment organisms is assessed. This data is interlinked with the assessment of bioaccumulation processes in this habitat.

B) option implementation

Without the prioritisation it is probable that comparatively weaker effects in soil and sediment remain undocumented next to the more obvious hazards to aqueous habitats. Bioaccumulation in the soil and in sediments might play an important role in creating a hazardous situation for organisms dwelling or growing in these habitats an over time.

Option 21 – Require considerations of most appropriate/relevant metrics

The appropriate metrics should be chosen also to develop and present a realistic exposure assessment and risk characterization. Mass metrics are not necessarily appropriate as a point of reference for the evaluation of nanomaterials.
4.7 Nano diamond

Applications
Nano diamonds are registered with REACH for a tonnage band of 1-10 t/y. According to communication with the FEPA the production range within Europe lies slightly above 1 t/y. After the initial discovery in the 1960s and the subsequent evolution of high throughput production processes in the 1990s the nano diamonds have found wide ranging applications in research and medicine as: substitutes for some quantum dots, nanoscale magnetic sensors, drug delivery platforms, surgical implant technology and medical instrument construction. In all of these fields the research and the subsequent medical implication is ongoing and picking up speed as more nano diamond becomes available. Similarly in industry nano diamonds find increasing application in tribology and lubrication, energy storage composites and catalysis.\(^\text{101}\) and citations therein

Physico chemical properties
Nano diamonds are also made of pure carbon but are polycrystalline in contrast to the monocrystalline conventional diamonds. They are translucent in appearance but consist of many granular crystals with a size range between 2-8 nm\(^\text{102}\) or 10-20 nm\(^\text{103}\) depending on the production process. The assortment of randomly oriented crystals conveys the exceptional mechanical properties that surpass the monocrystalline structure commonly associated with the term ‘diamond’. They are uniformly, exceptionally hard within a Knoop hardness range of 110-140 whereas the range of monocrystalline diamond is 60-120 on the same scale, due to the dependence on directionality of the applied pressure on a given crystal.\(^\text{103}\) The nano diamond is temperature stable up to 1200 °C in an inert atmosphere. The initial production process via chemical-vapour deposition (months) has been replaced since, first by high-pressure synthesis at 2300-2500 °C for a few minutes\(^\text{103}\) and later by an oxygen depleted TNT/hexogen (RDX) detonation.\(^\text{104}\)

Exposure
The majority of the nano diamonds are used in industrial applications where they are either used in closed systems (lubrication) with little to no chance for inhalation or as complex materials where a release of the nano diamonds is unlikely. The production of the nano diamonds requires complex technological setups that are also closed systems. The inhalation of the nano diamonds is overall unlikely outside of the scenario of an accident.

In research and medical context only low quantities are used and especially in the latter the nano diamonds are again either complexed or administered intravenously. Implants are set into place surgically. None of these applications show an obvious exposure hazard in light of the high biocompatibility nano diamonds are attributed with so far.

Environmental exposure is unlikely due to the highly specialized applications and the usage of nano diamonds within complexed materials in many cases. It has been estimated that nano diamonds

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\(^6\) Federation of European Producers of Abrasives: http://www.fepa-abrasives.org/DesktopDefault.aspx?portalname=&language=&folderindex=0&folderid=0&headingindex=0&headingid=0&tabindex=0&tabid=0
decrease the consumption of fossil fuels by ca. 5 % if they are added to lubricants used in motors. Here nano particles might be released into the environment via the exhaust fumes or during repair works or recycling.

As there are numerous possible modifications of the nano diamond surface the environmental distribution could affect soil and water compartments but there are no studies on the environmental distribution of nano diamonds up to now.

Environmental behaviour
Currently data on bioaccumulation and environmental distribution of nano diamonds is lacking.

Toxicology
Overall toxicity of nano diamonds is exceptionally low according to current research. Although lung overload still leads to inflammatory reactions of lung tissue the effects are reported to be muted in contrast to other nanomaterials in general and to carbon based materials in particular.\textsuperscript{96,105–108}

Some reports indicate low toxicity in dependence of the nano diamond source (i.e. surface modification or contamination from the manufacturer side) and of the acquired corona (see initial nanomaterial hazard statements).\textsuperscript{109}

Upon inhalation or instillation nano diamonds show no toxicity in the low dosage ranges 0,1-1 mg/kg and following that (0,8-20 mg/kg) a dosage dependent accumulation and pathology in lung, liver, kidney and blood.\textsuperscript{110,111} Neither long-term oral exposure of mice to nano diamonds nor subcutaneous exposure led to detectable pathologies or exhibited effects on the reproduction rate and the offspring.\textsuperscript{112,113} The genotoxicity results are contradictory and require further research.\textsuperscript{114,115}

There is no information available concerning environmental toxicity at this point but the test results for vertebrates indicate low toxicity. Further studies in invertebrates and bacteria are necessary to determine the environmental risk of nano diamonds.

Grading in the hazard table (see Final Report)
Nano diamonds are graded ‘low hazard’ in relation to other nanomaterials in this study because of the toxicology described above. Nano diamond is used as an example for nanomaterials of very low toxicity, even if in the case of nano diamond not all data is available to prove that. Although they show toxicity upon inhalation – as all nanomaterials do – the exhibited toxicity is comparably low. They are not water soluble and their distribution in the aquatic habitat, while possible from the physico-chemical characteristics, is not sufficiently studied yet. Judging from the physico-chemical properties little toxicity is expected.
4.8 Impact of the options on risk and exposure scenarios associated to nano diamond

The following tables illustrate where option implementation or lack thereof will affect the fields listed below without implying positive or negative nature of the impact. The tables serve as an orientation for the reader to illustrate those aspects that will be of particular interest in the following discussion and the ensuing upscaling of the impacts of the proposed modifications to REACH.

Table 4-5: Human health impacts of nano diamond

<table>
<thead>
<tr>
<th></th>
<th>options with impacts</th>
<th>pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>acute effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,11,12,21</td>
<td>local inflammation of the lung</td>
</tr>
<tr>
<td><strong>chronic effects</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>workers</td>
<td>6,12,21</td>
<td>fibrotic changes in the lung tissue</td>
</tr>
</tbody>
</table>

Table 4-6: Environmental impacts of nano diamond

<table>
<thead>
<tr>
<th></th>
<th>options with impacts</th>
<th>environmental impacts</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>accumulation/ persistency</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>biota</td>
<td>6,13,17,18,19,21</td>
<td>bioaccumulation</td>
</tr>
</tbody>
</table>
4.9 Impact of the option implementation to REACH on risk and exposure (nano diamond)

Option 6 – Include information on dustiness

A) status quo
The need for inhalation studies in the endpoint section is not apparent which has the potential to lead to incomplete exposure assessments. The implications for workers, consumers and ecological habitats are considerable as damage has to become apparent before further research is initialized. Furthermore this option specifies the need for these studies independent from the production or import quantities. Inhalation studies are otherwise required at a production/import tonnage of 10 t or more.

Without the data gained from inhalation study the need for specialized air filters and breathing masks at exposed workplaces might not become apparent.

B) option implementation
Nano diamonds are a dusty substance and risk assessments as well as necessary endpoint related studies are determined by this property. Inhalation studies with various size distributions and in different environments are necessary and the obtained data will be applied in security considerations along the lifecycle of products containing nano diamonds in inhalable form.

Option 11 – Require acute toxicity data for the most relevant route of exposure.
The toxicity data provided by the registrant should be coupled to the most relevant route of exposure of humans to a given substance and nanoform.

According to current knowledge nano diamond can be classified as non-hazardous and exposure assessments are thus not required.

Option 12 – Change particles to ‘(nano)particles’ for repeated dose toxicity studies (inhalation)
This option relates to the phrasing in the Annexes VIII, 8.6.1, IX, 8.6.2, and IUCLID section 8.5.2. The boundaries of the definitions here should be extended and specified by exchanging all instances of the word ‘particles’ for ‘(nano)particles’. Furthermore the following addition should be considered:
‘Especially for (nano)particles it should be justified when inhalation is not considered a relevant route of exposure.’

Either the test methods (for sub-acute, sub-chronic or combined repeated dose toxicity/reproductive screening study) or the guidance should refer to extended pathology/histology determinations and examination of relevant parameters in BAL (bronchoalveolar lavage) fluid.
A) status quo
Without the imposed fine resolution of the inhalation studies with regard to the employed size ranges of the nano particles and their modifications, little information is gained beyond the particular experimental setup. Specific applications and the alterations to the surface properties of the nano particles during the usage are the first ‘experiments’ indicating hazards to humans.

B) option implementation
This option will require the generation of inhalation specific toxicity data. The specific test methods dictated here will ensure good comparability to other materials.

Option 13 – Require non-bacterial, in vitro gene mutation study

A) status quo
If nano diamonds are genotoxic the circumstances of genotoxic activity might be dissimilar for bacteria and eukaryotes. Tests with living organisms are also often confounded by the fact that the secondary ROS production triggered by the inflammatory effects of nano particles in general can lead to gene deregulation. Without non-bacterial tests, it is impossible to assess potential genotoxicity inherent to nano diamonds in particular.

B) option implementation
Nano diamonds are not known to be genotoxic up to this point. It is unclear however if this is entirely valid until non-bacterial tests are documented. The results dictate the appropriate risk prevention measures.

Option 16 – Consider water solubility in relation to test waving
Nano particles are often not soluble in water. The lack of water solubility is not a reliable indicator for the lack of bioavailability. Nano particles have other options available to them to interact with their environment and biological cells.

A) status quo
Nano diamonds are not water soluble and thus testing of bioavailability, -accumulation, and persistence can be waived. But as it is also the case with other nano particles the lack of solubility is no deterrent for one of the three processes to occur. Without the necessary research done the risk assessment is likely to be imprecise.

B) option implementation
Nano diamonds are tested for all relevant endpoints and accurately classified for hazard and risk assessment within humans, other organisms, and the environment. As numerous results show the
lack of water solubility does not correlate with unavailability in the biosphere. This is especially true for nano particles.

**Option 17 – Specify that long term testing should not be waived based on lack of short term toxicity.**

Some nanoforms have a tendency to accumulate in biological systems and should thus not be discounted when considering toxicity just because they do not exhibit short term toxicity.

**A) status quo**

Nano diamonds exhibit remarkably low toxicity and mild, acute effects show only when comparatively large doses are administered. Using this as an indicator, further experiments for long term toxicity can be waived justifiably. Long term effects in humans or the environment might be overlooked depending on the manufacturer of the dossier.

**B) option implementation**

Nano diamonds are tested for long term pathological effects in humans and the environment. The information from such experiments is crucial for application specific risk assessment.

**Option 18 – Require that testing on soil and sediment organisms is prioritised**

As many nanomaterials are basically hydrophobic the current praxis of assessing the fate of nanomaterials in the environment on the base of equilibrium partitioning, could be reviewed. Especially sediments and soils are expected to be sinks for nanomaterials and the organisms living there should be given appropriate consideration, as the concentrations and modifications of the nanomaterials might be significantly higher and different than in the aqueous phase.

**A) status quo**

As nano diamonds are not soluble the deposition in soil and sediments is relevant but potentially overlooked. Bioaccumulation in the soil and in sediments might play an important role in creating a hazardous situation over time.

**B) option implementation**

The exposure characteristics of nano diamonds are easily altered by changing surface modifications in the course of the nano diamond lifecycle. The testing of soil and sediment organisms will quantify the amount of hazard in relation to surface modifications and in dependence of applications of the nano diamonds. The impact on soil and sediment organisms is assessed. This data is interlinked with the assessment of bioaccumulation processes in this habitat and adds to the complete profile of the nano diamonds for later risk assessment.
Option 19 – Specify that algae testing should not be waived based on insolubility

In accordance with the options 14-17 in this document algae testing should not be waived based on lack of water solubility.

As described above the lack of water solubility should not be a valid reason for the waiving of aquatic tests in the case of nanomaterials. Many are able to persist in aqueous media and influence their surroundings via agglomeration or surface modification with molecules found in the environment. This issue is also taken up by the ECHA in their new guidance document ECHA-12-G-05-EN.

See Option 16

Option 21 – Require considerations of most appropriate/relevant metrics

The appropriate metrics should be chosen also to develop and present a realistic exposure assessment and risk characterization.
5 Evaluation of knowledge gaps for seven representative nanomaterials due to the lack of the 21 options discussed above

5.1 Synthetic amorphous silica (SAS)

In 2009 the yearly production rate of nanoparticulate SiO$_2$ was estimated at 1.590.000 t (METI 2009$^{116}$). It is used mainly as high precision polishing agent, in plastics and rubber, paint, pigments, cosmetics, PET-bottles, drying agents and waterproofing spray. The surface area of the nano particles is highly dependent on the production technique. Exposure routes include oral ingestion of food or additives, dermal exposure via usage of cosmetics and toothpaste, and via inhalation of the nanoscale fraction of SiO$_2$ in concrete, rubber tires (abrasion), and paints.$^{117}$ As only very low amounts of most SiO$_2$ nano particles are water soluble the nano particles are mainly present in agglomerated form and tend to sediment quickly. If the agglomeration is hampered however the nano particles are difficult to separate from wastewater.$^{47}$ Inhalation of nano particles in high doses showed some pathological effects and the toxicity is inversely proportional to the particle size (basic risk of inhalable nanomaterial). High concentrations of nano particles in the aquatic compartment cause increased mortality rates and deformations within the local fauna.$^{47,63}$ The International Agency for Cancer Research (IARC) defines amorphous SiO$_2$ nano particle as not classifiable with regard to their carcinogenicity towards humans.$^{118}$ Currently SAS would be grouped somewhere between nano diamonds and TiO$_2$ nano particles because it is not classified as carcinogenic and does not by itself produce ROS. Nano diamonds however are not yet fully assessed with regard to their environmental impact. Thus SAS and nano diamonds might still find themselves grouped in the same hazard category. Due to different uses and surface chemistry exposures might vary considerably. In relation to the other nanomaterials in this report we group SAS nano particles in the low hazard category.

5.2 Carbon black

Carbon black is produced at an estimated tonnage of 8.100.000 t/y (ICBA$^7$). It is used mainly in the production of rubber (e.g. tires), as black pigment in paints, finishes and toner cartridges, in paper, plastics and fibres. Due to its electrical conductivity it is also used in electrodes and carbon brushes. It is water insoluble and is transferred into the environment mainly via tire abrasion, combustion processes and the usage of printers. In high concentrations it has been shown to be mutagenic, genotoxic, and potentially carcinogenic in humans. The presence of polycyclic aromatic hydrocarbons (PAHs) is unavoidable due to the production process but the PAHs do not contribute to the toxicity evaluations.$^{119,120}$ The IARC groups carbon black as possibly carcinogenic to humans (Group 2B).$^{43}$ There are further indications of negative effects on the blood circulatory system and it is suspected of aggravating allergic reactions. Aquatic organisms show very little effects upon exposure to high doses of carbon black. Because of the high doses necessary to observe the pathological effects and

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7 International Carbon Black Association (http://www.carbon-black.org/what_is.html)
the impurities in form of PAHs (attributed with the more toxic effects observed) present in previous experiments the danger to humanity and the environment is deemed low.\textsuperscript{52}

Due to the carcinogenic rating, the high production volumes, and the emphasis on toxicological effects on humans and not so much on the environment we group carbon black roughly with TiO\textsubscript{2} nano particles concerning the pathologic potential. As always the hazards detailed above have to be factored in with the exposures (heavily dependent on application, intentional and unintentional surface modifications).

5.3 Carbon nano tubes (CNTs) and multiwalled carbon nano tubes (MWCNTs)

An estimated amount of maximum 900 t of CNTs were produced per year in 2011.\textsuperscript{121} They are used in composite materials to mechanically strengthen them while adding little weight or/and to convey electrical conductivity. They are also used as antistatic coatings and are being investigated for the use as carriers for pharmacologically interesting molecules and substances. CNTs are mechanically resilient, can be single or multi-walled and display a high temperature conductivity while being water insoluble. In their pristine state they tend to agglomerate and accumulate at hydrophilic / hydrophobic interfaces. They are, however, easily modified by a multitude of organic molecules which convey new characteristics to them. Most commonly water solubility increases with modifications. Additional toxicological effects depend strongly on the type of modification. In the course of the production process they are also typically contaminated with trace metals from the catalysts which imbue them with toxic properties.\textsuperscript{52} Gustavsson et al. (2011)\textsuperscript{121} come to the conclusion that no direct evidence is available for carcinogenic effects of CNTs on humans but that this might be due to large information gaps. They find that “toxicological data is inadequate but indicates that there is a risk of imflammatory reaction and pulmonary fibrosis when inhaled at relatively low doses; there is also a risk of producing a DNA-damaging effect. They advise utmost precaution in production and handling facilities and the need to reduce the airborne exposure to a minimum. In addition to surface modifications and the composition of the nano tubes the aspect ratio is of critical importance when considering acute and chronic toxicity as well as the carcinogenic potential of CNTs. The higher the aspect ratio gets the higher the potential for frustrated phagocytosis and effects similar to those of carcinogenic asbestos fibres becomes. Besides length and aspect ratio rigidity plays another important role. Also here risk assessment is marred by the lack of knowledge and comparability between the many studies.\textsuperscript{122,123} The reason for the decision to include CNTs in this study is that production capacities about ten times higher than the current production volume are already available. Furthermore, due to the various fields of application that are currently investigated by the developers and producers, the production volume is very likely to increase significantly until 2022. If on-going research can be used as an indicator CNTs have the potential to find widespread application in many fields.

Here it is necessary to distinguish between workers exposed to CNTs or MWCNTs in the course of the production cycle and consumers which are usually exposed only to nano tubes tightly bound in the matrix of complex materials. Because some CNTs exhibit asbestos like characteristics upon inhalation they are graded with ‘medium hazard’. Nonetheless many CNT variations do not exhibit such properties and show only basic toxicology attributable to inhalable nanomaterials if they are not further modified. MWCNTs as they are mostly produced in Europe do not fulfil the fibre criterion of
the WHO by not being long or rigid enough to do so. The family of CNTs is highly diverse and a detailed analysis of all possible structural variations and aspect ratios would exceed the boundaries of the report.

5.4 Fullerenes

Fullerenes were produced at approximately 3 t/y worldwide in the year 2009.\textsuperscript{116} They have industrial and possibly medical applications. Industrially they are used as catalysts, in semi-conductors, as superconductors, base material for artificial diamonds, lubricants for high temperature applications, batteries, solar cells, fuel cells and sports devices. They are not dispersible in water in their pristine form just like the CNTs and the main exposure routes would be either via unskilled handling of the raw product or via their presence within cosmetic products (very few and not legally available on the European market). Though they are not toxic by themselves they show the same ability that CNTs have to accumulate other organic molecules on their surface. These convey additional characteristics to the Fullerene which can also include toxicity and the ability to penetrate the blood-brain barrier to unknown effect.\textsuperscript{52,124} For this reason Kahru et al. 2010 rated Fullerenes more hazardous than CNTs and TiO\textsubscript{2} nano particles.\textsuperscript{52} Although we agree with the inclusion of unintentional modifications into the risk assessment for nano particles we chose not to rate Fullerenes as toxic as Kahru et al. did as a consequence of an extensive literature search. The stochastic nature of the unintended nano particle surface modifications needs to impact on this assessment instead of focussing exclusively on toxic modifications. Regarding future production volumes, growth rates similar to CNT can be assumed. Hence, in analogy to CNT, Fullerenes have been selected as a case study despite of their relatively low current production volume. Large scale studies will be necessary to fully evaluate the risk posed by Fullerenes to human health as well as to the environment with an eye towards the exposure characteristics and the modifications imparted along the life cycle of specific applications.\textsuperscript{125}

Due to the possible variations of surface modifications within specific applications and along the life cycle Fullerenes are grouped with TiO\textsubscript{2} nano particles concerning the risk they pose. It is important however to keep in mind that the surface modifications are the crucial elements determining the hazard. In pristine state they show no enhanced toxicological properties and the potentially harmful surface modifications would hopefully be acquired unintentionally by the Fullerenes and thus be stochastic in nature. Lacking stronger, intrinsic toxicological properties and considering the other nanomaterials in this report we assign the relative grade ‘low hazard’ to Fullerenes.

5.5 Nano silver

In 2012, up to 550 t/y were produced.\textsuperscript{126} Nano silver is used as anti-microbial agent in textiles and packaging, medical products, cosmetic products, washing agents, paints and anti-biofouling coatings for marine vessels. Silver nano particles tend to aggregate as much as ZnO nano particles and the main toxicological effects stem from the ability to produce reactive oxygen species and the leaching of silver ions. The main exposure route is the loss of nano particles out of paints, finishes and textiles.\textsuperscript{74,127} Once in the waste water pathway, silver is oxidized to silver salts which have very low water solubility and therefore tend to accumulate in sediments and soil. Nano silver is rated as highly hazardous to water (water hazard class 3) but displays very little toxicity towards humans.\textsuperscript{52} On the one hand large knowledge gaps are still hindering conclusive risk assessment as also discussed by Christensen et al. and Johnston et al. (2010)\textsuperscript{128,129}, on the other hand many assessments point
towards the toxic properties of silver nano particles and still remain unheard due to non-scientific issues.\textsuperscript{130}

Due to the high toxicity towards aquatic habitats and the lack of data concerning many human health aspects we group nano silver with ZnO nano particles. The toxic effects mediated via ions similarly add to the toxic potential of both metal nano particles.

### 5.6 Nano copper

The yearly production of nano copper is between 1-10 t worldwide. It is widely used due to its cheap base material and its interesting industrial and end-user oriented applications. It is used in inks, lubricants, heat conductor fluids, coatings/catalysts, semiconductors and machine tools. It is also reported to be beneficial towards wound-healing\textsuperscript{131} and is found in some cosmetic products\textsuperscript{132}. The exposure to Cu nano particles can thus be via inhalation or via oral uptake. Inhalation of Cu nano particles leads to an increased probability for the occurrence of lung infections most probably due to the fact, that the neutrophil cells of the immune system are busy removing the nano particles and are thus less available for the immune defence against bacteria that cause the lung inflammation.\textsuperscript{133}

In this regard it is not clear whether Cu nano particles are more prone to facilitate lung infections than other nano particles. After all the involvement of the neutrophils in nano particle removal is universal so the added stress of invading bacteria would apply in any case of inhalatory exposure to nano particles. The inhalation of Cu nano particles does also trigger the mucociliary escalator which – also similar to any other inhaled nanomaterial – can lead to ingestion of the nanomaterial transported out of the lung. In comparison with many other metal oxides, Cu nano particles are very toxic upon ingestion. In spite of the fact that a daily uptake of Cu in the range of 70 and 100 µg/kg per adult human is normal ingestion of higher amounts of Cu quickly leads to heavy metal poisoning. This is mainly due to the Cu ions generated in the interaction between the gastric acid and the copper. As decreasing volume of a particle emphasizes the surface area disproportionately the smaller the Cu nano particles ingested are the larger the toxicological effects become.\textsuperscript{124} In case of Cu nano particles this increase in toxicity from bulk to nano form was quantified with a factor of 51.\textsuperscript{23} Besides the ion derived effects which affect mainly the lipid profile, pH values of blood and tissues, and tend to accumulate in renal tissue and the liver leading to dysfunction and cell death,\textsuperscript{135} the nano particles themselves are able to generate ROS. In spite of the fact that increasing surface oxidation and intentional modifications strongly diminish the amount of ROS produced the remaining ROS production is still sufficient to induce DNA damage and thus be carcinogenic.\textsuperscript{136}

Due to the increased toxicity of Cu nano particles vs. Cu bulk material on both pathways (ions and ROS generation) the Cu nano particles have been rated class 3 (moderately toxic) on the Hodge and Sterner Scale, while the bulk sized particles only warrant class 5 (practically non-toxic).\textsuperscript{135} Karlsson et al. (2008)\textsuperscript{137} classify Cu nano particles as the most toxic nano particles in comparison to TiO\textsubscript{2}-, ZnO-, Fe\textsubscript{2}-, and Fe\textsubscript{3}-nano particles in comparative inhalation studies. The same toxic effects described here can also be found in organisms of the aquatic habitat and Cu nano particles are known for their bactericidal and fungicidal properties.\textsuperscript{138,139}

Concerning their impact on human health when ingested orally the Cu nano particles should be grouped above the TiO\textsubscript{2} nano particles. There is little information on large scale environmental
effects but it can be expected that the negative ionic effects of Cu on the aquatic environment are within the same order of magnitude as ZnO ions. Cu salts are also grouped in the same category WGK 2 in Germany as ZnO is. Therefore we group Cu nano particles with ZnO and Ag nano particles in the category ‘high hazard’.

5.7 Quantum dots (e.g. Cadmium sulphide)

Remark: Quantum dots represent a diverse group of nanosized heavy metals by themselves and are listed here as representatives of a host of nanomaterials that are manufactured at less than 1 ton per year. Toxicological and exposure considerations will be exemplified using cadmium sulphide based particles.

In 2010 we estimate that less than 100 kg of quantum dots (QDs) were produced based on an average price range between 2500 and 6000 $ per gram and a market volume of ~67 mio $ in that year. Within this group several different types of nanomaterials are used. Due to their cost in production and the heavy metals contained their application range was limited to research and some LED applications. With the development of new QDs without heavy metals and the sinking prices QDs are investigated for the use in Electronics, Optoelectronics, Optics, and solar energy and a large growth of the market is predicted (BCC Research 2008). In their original and still widely applied incarnation QDs contain cadmium or selenium for example. Both metals display highly toxic properties in bacteria and mammals but in the context of quantum dots show highly diverging results concerning their toxicity. This is most probably due to extensive variations in their shell and outer coating compositions. The newer generation of QDs is designed without the need for a heavy metal core which might alleviate the toxicity significantly. With the redesign of the QDs their surface has also become hydrophilic thus opening new routes of exposure and ecological compartments that might be affected by their widespread usage.

Due to the usage of heavy metals in the original incarnation of QDs we grade these as highly hazardous.

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8 WGK: Wassergefährdungsklasse 2; water hazard class 2 denominates a substance that is considered hazardous to water dwelling organisms. In the German classification system there are three water hazard classes with class 3 being highly hazardous and class 1 weakly hazardous.
6 References


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