Nanotechnology Safety Resources

INTRODUCTION

The emerging field of nanotechnology has created an intense interest in the health risks associated with ultra-small matter. One nanometer is one-billionth of a meter.

These materials present new challenges in understanding, predicting, and managing potential health risks. The lack of data on nanoparticles makes protecting researchers from exposure a particular challenge. Neither do we know a great deal about potential health effects. To date, the data indicates:

Exposure to these materials during synthesis, manufacturing, and use may occur through inhalation, dermal contact, and ingestion.

Studies have indicated that low-solubility ultra fine particles are more toxic than larger particles on a mass for mass basis.

Because of their tiny size, they can get deep into the lungs and, once in the bloodstream, may be able to cross the blood-brain barrier. Particles deposited in the nasal region may be able to enter the brain by translocation along the olfactory nerve. Particles, especially composites, may be able to penetrate the skin, and even some protective equipment. The smaller the particle, the more likely it is to be suspended in air, and hence, be available for inhalation/ingestions.

Other hazards to consider are catalytic effects and fire or explosion.

Because of the limited information on the risks of handling these materials, workers should implement stringent controls on exposure when working with them. Until more knowledge becomes available, EH&S is, for now, providing the following guidelines and references for more information.

Check back every 3 months for updates and changes. Please fell free to contact Debbie-Wolfe Lopez in EHS at 404-385-2964 if you have questions

Recommended Safety Procedures for handling nanomaterials

1. Use good general laboratory safety practices as found in the Laboratory Safety Manual, the Bio-Safety Manual and individual laboratory operating procedures. Wear gloves, lab coats, safety glasses, face shields, closed-toed shoes. Avoid getting nanoparticles in eyes, mucous membranes, on skin, or in respiratory tract.
2. Wash your hands BEFORE you leave the lab.
3. Be sure to consider the hazards of precursor materials in evaluating process hazards. (for example, some powders are more dangerous until they are mixed into a solution, whereby they become safer to handle – less possibility of inhalation of floating particles.)
4. Avoid skin contact with nanoparticles or nanoparticle-containing solutions by using appropriate

personal protective equipment. Do not handle nanoparticles with your bare skin.

5. Handle nanoparticles only inside a HEPA-filtered powered-exhaust laminar flow hood, wear appropriate respiratory protection. If this is not possible, consult with EH&S on obtaining respiratory protection.
6. Use fume exhaust hoods to expel fumes from tube furnaces or chemical reaction vessels.
7. Waste that contains nanoparticles should be placed in puncture proof sealable containers, or double bagged in 6 ml plastic, clearly marked with contents and disposed of through hazardous waste channels.
8. Clean up of spilled nanoparticles should be accomplished with a HEPA filtered vacuum or call EH&S. Filters from such vacuums are to be disposed of as in 6, above.
9. Equipment , facilities, and supplies used for nano-research, must be evaluated prior to removal, maintenance, or conversion to other uses.
10. Become familiar with the MSDS associated with the basic material; be alert for the onset of any symptoms associated with the chronic effects of these materials.
11. No one set of rules will cover all situations. EH&S strongly recommends that you consult with us on any particles project BEFORE project initiation.
12. Given the differing synthetic methods and experimental goals, no blanket recommendation can be made regarding aerosol emissions controls. This should be evaluated on a case by case basis.
13. Consideration should be given to the high reactivity of some nanopowders materials with regard to potential fire and explosion hazards.

- National Institute for Occupational Safety and Health

NIOSH Safety and Health Topic:

Nanotechnology

http://www.cdc.gov/niosh/topics/nanotech/safenano/healthconcerns.html

Potential Health Concerns

Nanotechnology is an emerging field. As such, there are many uncertainties as to whether the unique properties of engineered nanomaterials (which underpin their commercial potential) also pose occupational health risks. These uncertainties arise because of gaps in knowledge about the factors that are essential for predicting health risks—factors such as routes of exposure, translocation of materials once they enter the body, and interaction of the materials with the body's biological systems. The potential health risk following exposure to a substance is generally associated with the magnitude and duration of the exposure, the persistence of the material in the body, the inherent toxicity of the material, and the susceptibility or health status of the person. More data are needed on the health risks associated with exposure to engineered nanomaterials. Results of existing studies in animals or humans on exposure and response to ultrafine or other respirable particles provide a basis for preliminary estimates of the possible adverse health effects from exposures to similar engineered materials on a nano-scale. Experimental studies in rodents and cell cultures have shown that the toxicity of ultrafine or nanoparticles is greater than that of the same mass of larger particles of similar chemical composition [Oberdörster et al., 1992, 1994a,b; Lison et al., 1997; Tran et al., 1999, 2000; Brown et al., 2001; Duffin et al., 2002; Barlow et al. 2005]. In addition to particle surface area, other particle characteristics may influence the toxicity, including solubility, shape, and surface chemistry [Duffin et al. 2002; Oberdörster et al. 2005a; Maynard and Kuempel 2005; Donaldson et al. 2006]. More research is needed on the influence of particle properties on interactions with biological systems and the potential for adverse effects. International research strategies for evaluating the safety of nanomaterials are actively being developed through cooperative efforts [Thomas et al. 2006].

Existing toxicity information about a given material can also help provide a baseline for anticipating the possible adverse health effects that may occur from exposure to that same material on a nanoscale.

A. Exposure Routes

The most common route of exposure to airborne particles in the workplace is by inhalation. The deposition of discrete nanoparticles in the respiratory tract is determined by the particle's aerodynamic or thermodynamic diameter (depending on particle size). Agglomerates of nanoparticles will deposit according to the diameter of the agglomerate, not constituent nanoparticles. Research is still ongoing to determine the physical factors that contribute to the agglomeration and de-agglomeration of nanoparticles, and the role of agglomerates in the toxicity of inhaled nanoparticles.

Discrete nanoparticles are deposited in the lungs to a greater extent than larger respirable particles [ICRP 1994], and deposition increases with exercise due to increase in breathing rate and change from nasal to mouth breathing[Jaques and Kim 2000; Daigle et al. 2003] and among persons with existing lung diseases or conditions [Brown et al. 2002]. Based on animal studies, discrete nanoparticles may enter the bloodstream from the lungs and translocate to other organs [Takenaka et al. 2001; Nemmar et al. 2002; Oberdörster et al. 2002].

Discrete nanoparticles (35-37 nm count median diameter) that deposit in the nasal region may be able to enter the brain by translocation along the olfactory nerve, as was recently observed in rats [Oberdörster et al. 2004; Oberdörster et al. 2005a]. The transport of insoluble particles from 20 to 500 nm diameter to the brain via sensory nerves (including olfactory and trigeminus) was reported in earlier studies in several animal models [De Lorenzo 1970; Adams and Bray 1983; Hunter and Dey 1998]. This exposure route has not been studied in humans, and research is continuing to evaluate its relevance.

Ingestion is another route whereby nanoparticles may enter the body. Ingestion can occur from unintentional hand to mouth transfer of materials; this can occur with traditional materials, and it is scientifically reasonable to assume that it also could happen during handling of materials that contain nanoparticles. Ingestion may also accompany inhalation exposure because particles that are cleared from the respiratory tract via the mucociliary escalator may be swallowed [ICRP 1994]. Little is known about possible adverse effects from the ingestion of nanoparticles.

Some studies suggest that nanoparticles also could enter the body through the skin during occupational exposure. The U.K. Royal Society and Royal Academy of Engineers have reported that unpublished studies indicate nanoparticles of titanium dioxide used in sunscreens do not penetrate beyond the epidermis [The Royal Society and The Royal Academy of Engineering 2004]. However, the report also makes a number of recommendations addressing the need for further and more transparent information in the area of nanoparticle dermal penetration. Tinkle *et al. [2003]* have shown that particles smaller than 1 µm in diameter may penetrate into mechanically flexed skin samples.

A more recent study reported that nanoparticles with varying physicochemical properties were able to penetrate the intact skin of pigs (Ryman-Rasmussen et al. 2006). These nanoparticles were quantum dots of different size, shape, and surface coatings. They were reported to penetrate the stratum corenum barrier by passive diffusion and localize within the epidermal and dermal layers within 8 to 24 hours. The dosing solutions were two- to four-fold dilutions of quantum dots as commercially supplied and thus represent occupationally relevant doses. This study suggests that the skin is a potential route of exposure for nanoparticles.

At this time, it is not known if skin penetration of nanoparticles would result in adverse effects as these studies have not been reported in animal models. Studies conducted in vitro using primary or cultured human skin cells have shown that both SWCNT and multi-walled carbon nanotubes (MWCNT) can enter cells and cause release of pro-inflammatory cytokines, oxidative stress, and decreased viability [Monteiro-Riviere et al. 2005; Shvedova et al. 2003]. It remains unclear, however, how these findings may be extrapolated to a potential occupational risk, given that additional data are not yet available for comparing the cell model studies with actual conditions of occupational exposure. Research on the dermal exposure of nanoparticles is ongoing [www.uni-leipzig.de/~nanoderm/].

B. Effects Seen in Animal Studies

Experimental studies in rats have shown that at equivalent mass doses, insoluble ultrafine particles are more potent than larger particles of similar composition in causing pulmonary inflammation, tissue damage, and lung tumors [Lee et al. 1985; Oberdörster and Yu 1990; Oberdörster et al. 1992, 1994a,b; Heinrich et al. 1995; Driscoll 1996; Lison et al. 1997; Tran et al. 1999, 2000; Brown et al. 2001; Duffin et al. 2002; Renwick et al. 2004; Barlow et al. 2005]. These studies have shown that for poorly-soluble and low toxicity (PSLT) particles, the dose-response relationships are consistent across particle sizes when dose is expressed as particle surface area. In addition to particle size and surface area, studies have also shown that other particle characteristics can influence toxicity. For example, although the relationship between particle surface area dose and pulmonary inflammation is consistent among PSLT particles, crystalline silica is much more inflammogenic than PSLT particles at a given surface area dose [Duffin et al. 2002].

These studies indicate that for nanoparticles with similar properties (e.g., PSLT) , the toxicity of a given mass dose will increase with decreasing particle size due to the increasing surface area. However, the dose-response relationship may differ for particles with different chemical composition and other properties. Consistent with these findings, a recent study reported doses of either fine or ultrafine TiO2 in rats at which the lung responses did not significantly differ from controls, while crystalline silica caused more severe

lung responses at the same dose [Warheit et al. 2006]. That study was unable to adequately test hypotheses about particle surface area dose and toxicity because the rat lung responses to either fine or ultrafine TiO2 did not significantly differ from controls.

PTFE Fume

Among ultrafine particles, freshly-generated polytetrafluoroethylene (PTFE) fume (generated at temperatures >425oC) is known to be highly toxic to the lungs. Freshly-generated PTFE fume caused hemorrhagic pulmonary edema and death in rats exposed to less than 60 µg/m3 [Oberdörster et al. 1995]. In contrast, aged PTFE fume was much less toxic and did not result in mortality, which was attributed to the increase in particle size from accumulation and to changes in surface chemistry [Johnston et al. 2000; Oberdörster et al. 2005a]. Human case studies have reported pulmonary edema in workers exposed to PTFE fume and an accidental death in a worker when an equipment malfunction caused overheating of the PTFE resin and release of the PTFE pyrolysis products in the workplace [Goldstein et al. 1987; Lee et al. 1997]. While PTFE fume differs from engineered nanoparticles, these studies illustrate properties of ultrafine particles that have been associated with an acute toxic hazard. Enclosed processes and other engineering controls appear to have been effective at eliminating worker exposures to PTFE fume in normal operations, and thus may provide examples of control systems that may be implemented to prevent exposure to nanoparticles that may have similar properties.

Carbon nanotubes

Carbon nanotubes (CTN) are specialized forms or structures of engineered nanoparticles that have had increasing production and use [Donaldson et al. 2006]. Consequently, a number of toxicological studies of CNT have been performed in recent years. These studies have shown that the toxicity of CNT may differ from that of other nanoparticles of similar chemical composition. For example, single-walled CNTs (SWCNT) have been shown to produce adverse effects including granulomas in the lungs of mice and rats at mass doses of carbon that did not produce these adverse effects [Shvedova et al. 2005; Lam et al. 2004]. While both SWCNTs and carbon black are carbon-based, SWCNTs have a unique convoluted fibrous structure and specific surface chemistry that offers excellent electrical conductive properties. How these characteristics may influence toxicity is not known. CNTs may contain metal catalysts as byproducts of their production, which could also contribute to their toxicity.

In a study of SWCNTs instilled into the lungs of rats, multi-focal granulomas (without transient inflammation or persistent lesions) were observed at doses of 1 or 5 mg/kg body weight [Warheit et al. 2004]. In a study of mice instilled with one of several types of SWCNTs (raw, purified, iron-containing, and nickel-containing) at doses of 0.1 or 0.5 mg/mouse (approximately 3 or 16 mg/kg body weight), dose-dependent epithelioid granulomas were observed at 7 days, which persisted at 90 days [Lam et al. 2004,2006]. Both the raw and purified forms produced interstitial inflammation, while mortality (5/9 mice) was observed in the high dose group of the Ni-containing SWCNT.

NIOSH researchers recently reported adverse lung effects following pharyngeal aspiration of SWCNTs in mice using doses between 10-40 µg/mouse (approximately 0.5–2 mg/kg body weight) [Shvedova et al. 2005]. The findings showed that exposure to SWCNTs in mice lead to transient pulmonary inflammation, oxidative stress, decrease in pulmonary function, decrease in bacterial clearance, and early onset of interstitial fibrosis. Deposition of agglomerates resulted in development of granulomas, while deposition of more dispersed nanotube structures resulted in the rapid development of interstitial fibrosis (within 7 days), which progressed over a 60 day post-exposure period.

SWCNT was more fibrogenic than an equal mass of either ultrafine carbon black or fine quartz [Shvedova et al. 2005; Lam et al. 2004]. Based on their findings in mice, Shvedova et al. [2005] estimated that workers may be at risk of developing lung lesions if they were exposed to SWCNT over a period of 20 days at the current OSHA Permissible Exposure Limit (PEL) for graphite (5 mg/m3). Lam et al. [2004, 2006] provided similar estimates and suggested that the graphite PEL should not be used (e.g., on MSDS) as a safe concentration for workers exposed to CNTs. Compared to instillation, the pharyngeal aspiration technique may approximate more closely the particle deposition that occurs during inhalation, although inhalation studies of CNTs may provide more definitive information about their potential toxicity in humans [Donaldson et al. 2006].

Multi-walled CNTs (MWCNT) were recently studied by intratracheal instillation in Sprague-Dawley rats receiving either 0.5, 2, or 5 mg (approximately 2, 9, or 22 mg/kg body weight) of either ground MWCNT or unground MWCNT [Muller et al. 2005]. Both forms produced pulmonary inflammation and fibrosis. The dispersion in the lungs was greater for the ground MWCNT, and fibrotic lesions were observed in the deep lungs (alveolar region) of the ground MWCNT-treated rats, while fibrosis was primarily seen in the airways of the rats treated with unground MWCNT. The biopersistence of the unground CNT was greater than that of the ground MWCNT, with 81% vs. 36%, respectively, remaining in the lungs at day 60. At an equal mass dose, ground MWCNT produced a similar inflammatory and fibrogenic response as chrysotile asbestos and a greater response than ultrafine carbon black [Muller et al. 2005]. Ground CNTs are used in polymer composites and other matrixes, and thus there is a potential for worker exposure to either ground or unground CNT.

These studies indicate the need for more data on potential exposures of workers to CNTs. Maynard et al. [2004] reported relatively low airborne mass concentrations of raw SWCNT material in one facility, although concentrations increased considerably when the material was agitated. Given the unusual toxicity of SWCNT observed in rodent lungs at relatively low mass doses and the uncertainty about potential adverse effects in workers if exposed, it is prudent to minimize worker exposure to airborne CNTs through the use of effective engineering controls, work practices, and personal protective equipment (see Section on Exposure Control Procedures)

C. Observations from Epidemiological Studies Involving Fine and Ultrafine Particles

Epidemiological studies in workers exposed to aerosols including fine and ultrafine particles have reported lung function decrements, adverse respiratory symptoms, chronic obstructive pulmonary disease, and fibrosis [Kreiss et al. 1997; Gardiner et al. 2001; Antonini 2003]. In addition, some studies have found elevated lung cancer among workers exposed to certain ultrafine particles, e.g., diesel exhaust particulate [Steenland et al. 1998; Garshick et al. 2004] or welding fumes [Antonini 2003]. The implications of these studies to engineered nanoparticles, which may have different particle properties, are uncertain.

Epidemiological studies in the general population have shown associations between particulate air pollution and increased morbidity and mortality from respiratory and cardiovascular diseases [Dockery et al. 1993; HEI 2000; Pope et al. 2002; Pope et al. 2004]. Some epidemiological studies have shown adverse health effects associated with exposure to the ultrafine particulate fraction of air pollution [Peters et al. 1997; Penttinen et al. 2001; Ibald-Mulli et al. 2002; Timonen et al. 2004; Ruckerl et al. 2005], although uncertainty exists about the role of ultrafine particles relative to the other air pollutants in causing the observed adverse health effects. The associations in these studies have been based on measurements of the particle number or mass concentrations of particles within certain size fractions (e.g., PM2.5). In an experimental study of healthy and asthmatic subjects inhaling ultrafine carbon particles, changes were observed in the expression of adhesion molecules by blood leukocyte, which may relate to possible cardiovascular effects of ultrafine particle exposure [Frampton et al. 2006].

D. Hypotheses from Animal and Epidemiological Studies

The existing literature on particles and fibers provides a scientific basis from which to evaluate the potential hazards of engineered nanoparticles. While the properties of engineered nanoparticles can vary widely, the basic physicochemical and toxicokinetic principles learned from the existing studies are relevant to understanding the potential toxicity of nanoparticles. For example, we know from studies in humans that a greater proportion of inhaled nanoparticles will deposit in the respiratory tract (both at rest and with exercise) compared to larger particles [ICRP 1994; Jaques and Kim 2000; Daigle et al. 2003; Kim and Jaques 2004]. We know from studies in animals that nanoparticles in the lungs can be translocated to other organs in the body, although it is not well known how this may be influenced by the chemical and physical properties of the nanoparticles [Takenaka et al. 2001; Kreyling et al. 2002; Oberdörster et al. 2002, 2004; Semmler et al. 2004; Geiser et al. 2005]. Due to their small size, nanoparticles can cross cell membranes and interact with subcellular structures such as mitochondria, where they have been shown to cause oxidative damage and impair function of cells in culture [Möller et al. 2002, 2005; Li et al. 2003; Geiser et al. 2005]. Animal studies have shown that nanoparticles are more biologically active due to their greater surface area per mass compared with larger-sized particles of the same chemistry [Oberdörster et al. 1992; 1994a,b; 2005a; Driscoll 1996; Lison et al. 1997; Brown et al. 2001; Duffin et al. 2002; Renwick et al. 2004]; Barlow et al. 2005]. While this increased biological activity of nanoparticles is a fundamental component to the utility of nanoparticles for industrial, commercial, and medical applications, the consequences of unintentional exposures of workers to nanoparticles are uncertain.

Research reported from laboratory animal studies and from human epidemiological studies have lead to hypotheses regarding the potential adverse health effects of engineered nanoparticles. These hypotheses are based on the scientific literature of particle exposures in animals and humans. This literature has been recently reviewed [Donaldson et al. 2005; Maynard and Kuempel 2005; Oberdörster et al. 2005a, Donaldson et al. 2006]. In general, the particles used in past studies have not been characterized to the extent recommended for new studies in order to more fully understand the particle properties influencing toxicity [Oberdörster et al. 2005b; Thomas et al. 2006]. As this research continues, more data will become available to support or refute these hypotheses for engineered nanoparticles.

1. Exposure to engineered nanoparticles is likely to cause adverse health effects similar to well-characterized ultrafine particles that have similar physical and chemical characteristics.

Studies in rodents and humans support the hypothesis that exposure to incidental ultrafine particles pose a greater respiratory hazard than the same mass of larger particles with a similar chemical composition. Studies of existing particles have shown adverse health effects in workers exposed to ultrafine particles (e.g., diesel exhaust particulate, welding fumes), and animal studies have shown that ultrafine particles are more inflammogenic and tumorigenic in the lungs of rats than an equal mass of larger particles of similar composition [Oberdörster and Yu 1990; Driscoll 1996; Tran et al. 1999, 2000]. **If engineered nanoparticles have the same physiochemical characteristics that are associated with reported effects from ultrafine particles, they may also pose the same health concerns.**

Although the physiochemical characteristics of existing ultrafine particles and engineered nanoparticles can differ substantially, the toxicological and dosimetric principles derived from available studies may be relevant to postulating the health concerns for new engineered particles. The biological mechanisms of particle-related lung diseases (e.g., oxidative stress, inflammation, and production of cytokines, chemokines, and cell growth factors) [Mossman and Churg 1998; Castranova 2000, Donaldson and Tran 2002] appear to be a consistent lung response for respirable particles including ultrafine or nanoparticles [Donaldsonet al. 1998; Donaldson and Stone 2003; Oberdörster et al. 2005]. Toxicological studies have shown that the chemical and physical properties that are important factors

influencing the fate and toxicity of ultrafine particles may also be significant for other nanoparticles [Duffin et al. 2002; Kreyling et al. 2002; Oberdörster et al. 2002; Semmler et al. 2004].

2. Surface area and activity, particle number may be better predictors of potential hazard than mass.

The greater potential hazard may relate to the greater number or surface area of nanoparticles compared with that for the same mass concentration of larger particles [Oberdörster et al. 1992; Oberdörster et al. 1994a,b; Driscoll et al. 1996; Tran et al. 2000; Brown et al. 2001; Peters et al. 1997; Moshammer and Neuberger 2003]. This hypothesis is based primarily on the pulmonary effects observed in studies of rodents exposed to various types of ultrafine or fine particles (e.g., titanium dioxide, carbon black, barium sulfate, carbon black, diesel soot, coal fly ash, and toner) and in humans exposed to aerosols including nanoparticles (e.g., diesel exhaust and welding fumes). These studies indicate that for a given mass of particles, relatively insoluble nanoparticles are more toxic than larger particles of similar chemical composition and surface properties. Studies of fine and ultrafine particles have shown that particles with less reactive surfaces are less toxic [Tran et al. 1999; Duffin et al. 2002].

However, even particles with low inherent toxicity (e.g., titanium dioxide) havebeen shown to cause pulmonary inflammation, tissue damage, and fibrosis at sufficiently high particle surface area doses [Oberdörster et al. 1992, 1994 a,b; Tran et al. 1999, 2000].

Through engineering, the properties of nanomaterials can be modified. For example, a recent study has shown that the cytotoxicity of water-soluble fullerenes can be reduced by several orders of magnitude by modifying the structure of the fullerene molecules (e.g., by hydroxylation) [Sayes et al. 2004]. These structural modifications were shown to reduce the cytotoxicity by reducing the generation of oxygen radicals – which is a probable mechanism by which cell membrane damage and death occurred in these cell cultures. Increasing the sidewall functionalization of SWCNT also rendered these nanomaterials less cytotoxic to cells in culture [Sayes et al. 2005]. Cytotoxicity studies with quantum dots have shown that the type of surface coating can have a significant effect on cell motility and viability [Hoshino et al. 2004; Shiohara et al. 2004; Lovric et al. 2005]. Differences in the phase composition of nanocrystalline structures can influence their cytotoxicity; in a recent study comparing two types of titanium dioxide nanoparticles, anatase was more cytotoxic and produced more reactive species than did rutile with similar specific surface area (153 and 123 m2g, respectively) [Sayes et al. 2006]. Reactive oxygen species were also associated with the cytotoxicity of titanium dioxide nanoparticles to mouse microglia (brain cells) grown in culture [Long et al. 2006].

The studies of ultrafine particles may provide useful data to develop preliminary hazard or risk assessments and to generate hypotheses for further testing. The studies in cell cultures provide information about the cytotoxic properties of nanomaterials that can guide further research and toxicity testing in whole organisms. More research is needed of the specific particle properties and other factors that influence the toxicity and disease development associated with airborne particles, including those characteristics that may be most predictive of the potential safety or toxicity of new engineered nanoparticles.

http://www.cdc.gov/niosh/topics/nanotech/safenano/healthconcerns.html

http://www.cdc.gov/niosh/topics/nanotech/safenano/safetyhazards.html

Other helpful sites include:

- ACS Chemical & Engineering News Nanotechnology website
- ACS Chemical & Engineering News Article: Nano Database Goes Online
- American National Standard's Nanotechnology Standards Panel
- Center for Responsible Nanotechnology
- The Royal Society and Royal Academy of Engineering
- National Nanotechnology Initiative Environment and Health Safety Research
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