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Nanotechnologies — Health and safety practices in occupational settings relevant to nanotechnologies

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TECHNICAL REPORT

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Nanotechnologies — Health and safety practices in occupational settings relevant to nanotechnologies

Nanotechnologies — Pratiques de sécurité dans les arrangements professionnels relatifs aux nanotechnologies

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Foreword

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In exceptional circumstances, when a technical committee has collected data of a different kind from that which is normally published as an International Standard ("state of the art", for example), it may decide by a simple majority vote of its participating members to publish a Technical Report. A Technical Report is entirely informative in nature and does not have to be reviewed until the data it provides are considered to be no longer valid or useful.

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1. Introduction

The field of nanotechnologies is advancing rapidly and is expected to impact virtually every facet of global industry and society. International standardization on nanotechnologies should contribute to realizing the potential of this technology for the betterment and sustainability of our world through economic development, improving the quality of life, and for improving and protecting public health and the environment. One can expect many new engineered nanomaterials coming to the market place and work place. The introduction of these new materials into the workplace raises questions concerning occupational safety and health that should be addressed, as appropriate, by international standards. While such standards are being developed, it is important, through this Technical Report, to assemble and make available to users, useful knowledge on occupational safety and health practices in the context of nanotechnologies.

Nanotechnology involves materials at the nanoscale. As a working definition,[i] the "nanoscale" means size range from approximately 1 nm to 100 nm. A nanometer is 1×10^{-9} m or one millionth of a millimeter. It is difficult to fully appreciate these remarkably small scales. To give a sense of this scale, a human hair is of the order of 10,000 to 100,000 nm, a single red blood cell has a diameter of around 5,000 nm, viruses typically have a maximum dimension of 10 to 100 nm and a DNA molecule has a diameter of around 2 nm. The term "nanotechnology" can be misleading since it is not a single technology or scientific discipline. Rather it is a multidisciplinary grouping of physical, chemical, biological, engineering, and electronic processes, materials, applications and concepts in which the defining characteristic is one of size.

The distinctive and often unique properties which are observed with nanomaterials offer the promise of broad advances for a wide range of technologies in fields as diverse as computers, biomedicine, and energy. At this early stage the potential applications of nanomaterials seem to be limited only by the imagination. Articles appear daily in the scientific and popular press and on a host of websites dedicated to the field. New companies, often spin outs from university research departments, are being formed and are finding no shortage of investors willing to back their ideas and products. New materials are being discovered or produced and astonishing claims are being made concerning their properties, behaviors and applications. As of June, 2007, over 400 nano-enabled new products are listed in an inventory of products already utilizing nanotechnology compiled by the Woodrow Wilson Center's Project on Emerging Nanotechnologies (www.nanotechproject.org/inventories/consumer/). Another list of products can also be found on U. S. National Nanotechnology Initiative web-site at www.nano.gov/html/facts/appsprod.html. While much of the current "hype" is highly speculative, there is no doubt that worldwide, governments and major industrial companies are committing significant resources for research into the development of nanometer scale processes, materials and products.

Ordinary materials such as carbon or silicon, when reduced to the nanoscale, often exhibit novel and unexpected characteristics such as extraordinary strength, chemical reactivity, electrical conductivity, or other characteristics that the same material does not possess at the micro or macro-scale. A huge range of nanomaterials have already been produced including nanotubes, nanowires, fullerene derivatives (bucky balls).

A few engineered nanomaterials were developed already in the 19th and 20th centuries, at a time when the word "nanotechnology" was unknown. Among such nanomaterials are zeolites, catalyst supports such as $MgCl_2$, pigments and active fillers such as carbon black and synthetic amorphous silica. Market size of these commodity materials is well above the billion US dollars or million tons threshold.

Nanotechnologies are gaining in new commercial application. Nanomaterials are currently being used in electronic, magnetic and optoelectronic, biomedical, pharmaceutical, cosmetic, energy, catalytic and materials applications. Areas producing the greatest revenue for nanomaterials are chemical-mechanical polishing, magnetic recording tapes, sunscreens, automotive catalyst supports, electro-conductive coatings and optical fibers.

[i] Please note, that definitions used throughout this Technical Report are based on draft definitions developed by ISO TC 229 WG1 and might become obsolete if draft definitions change.

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The occupational health and safety effects of new nanomaterials are mostly unknown. This can be attributed to the relatively recent development of the nanotechnology sector and, as a result, the lack of available information on human exposures and working conditions. As a consequence our abilities to accurately predict the impact of some nanomaterials exposures on worker health are limited at this time. In particular our abilities to measure nanoparticles in the workplace (or more generally) are limited by current technologies. Nanotechnology presents us with new challenges as the properties of nanomaterials now depend on size and shape as much as the more conventional factors of chemical structure and composition. Measuring these additional attributes will be necessary to accurately assess nanomaterials in the workplace. In addition, the capability of the human body to recognize and appropriately respond to most nanomaterials is essentially unknown at the moment. On the other hand, in the case of some nanostructured materials, such as carbon black and synthetic amorphous silica, toxicologic and epidemiologic data are available.

There are many gaps in current science about identifying, characterizing, and evaluating potential occupational exposures in the nanotechnology context. These gaps in our knowledge will best be addressed at a multidisciplinary level. Occupational health practitioners and scientists and practitioners in the toxicology field including medical scientists and environmental scientists have vital roles to play in safeguarding health in this fast-moving field. Collaborative studies - ideally with international coordination - are essential in order to provide the critical information required within a reasonable time frame.

2. Scope

This Technical Report describes health and safety practices in occupational settings relevant to nanotechnologies. The initial outline was prepared using U. S. NIOSH's Approaches to Safe Nanotechnology: An Information Exchange with NIOSH.[1] This Technical Report focuses on the occupational manufacture and use of engineered nanomaterials. It does not address health and safety issues or practices associated with nanomaterials generated by natural processes, hot processes and other standard operations which unintentionally generate nanomaterials, or potential consumer exposures or uses, though some of the information in this Technical Report might be relevant to those areas. For more general information on the environment, health and safety of nanotechnologies, the reader can refer to other existing well documented reviews.[2-7] Use of the information in this Technical Report could help companies, researchers, workers and other people to prevent adverse health and safety consequences during the production, handling, use and disposal of manufactured nanomaterials. This advice is broadly applicable across a range of nanomaterials and applications.

This Technical Report is based on current information about nanotechnologies, including characterization, health effects, exposure assessments, and control practices. The authors of the Technical Report have attempted to remain current with the use of terms and their definitions. However, definitions in this field are evolving and some terms have not yet undergone ISO consensus review. Therefore, the terms are intended to be used solely for the purpose of this Technical Report and not to be considered formal definitions beyond this Technical Report. It is expected that this Technical Report will be revised and updated and new safety standards will be developed as our knowledge increases and experience is gained in the course of technological advance.

Bibliography

[1] U. S. NIOSH, Approaches to Safe Nanotechnology: An Information Exchange with NIOSH, 2006. Available online at: http://www.cdc.gov/niosh/topics/nanotech/safenano/. (Accessed on July 23, 2007).

[2] Royal Society/Royal Academy, Nanoscience and nanotechnologies: Opportunities and uncertainties, 2004.

[3] U. S. NIOSH, Strategic plan for NIOSH nanotechnology research filling the knowledge gaps, 2005. Available on line at http://www.cdc.gov/niosh/topics/nanotech/strat_planINTRO.html. (Accessed on July 23, 2007).

[4] ILSI, Principles for characterizing the potential human health effects from exposure to nanomaterials: Elements of a screening strategy, 2005.

[5] SCENIHR, Opinion on the appropriateness of existing methodologies to assess the potential risks associated with engineered and adventitious products of nanotechnologies, 2007.

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[6] U. S. EPA, Nanotechnology white paper, 2007. Available at http://www.epa.gov/OSA/nanotech.htm. (Accessed on July 23, 2007).

[7] U. S. NIOSH, Progress toward safe nanotechnology in the workplace, NIOSH Publication No. 2007-123, 2007. Available on line at http://www.cdc.gov/niosh/docs/2007-123/. (Accessed on July 23, 2007).

3. Nanomaterials: description and manufacturing

3.1. Engineered nanomaterials

Engineered nanomaterials are designed with specific properties in mind. Engineered nanomaterials encompass nano-objects and nanostructured materials. The former are defined as materials with one (nanoplate), two (nanorod) or three external dimensions (nanoparticle) in the nanoscale (i.e. between approximately 1 and 100 nm). Examples of nanostructured materials are nanocomposites composed of nano-objects embedded in a solid matrix or nano-objects bonded together in simple random assemblies as in aggregates and agglomerates or ordered as in crystals of fullerenes or carbon nanotubes.[1] Discussion in this Technical Report will focus primarily on nano-objects and their simple assemblies.

Relatively simple nanomaterials presently in use or under active development can be classified in terms of dimensionality and the primary chemical composition. However, even simple nanomaterials are often coated and have complex chemical and physical structure. Any attempt to classify nanomaterials is highly artificial with many materials falling into several classification categories. Thus, the following description is for organizational purposes only.

Quantum dots and fullerenes are confined to the three-dimensional nanoscale domain. Nanotubes, nanowires, nanofibers and nanofibrils have at least two nanoscale dimensions, while nanoscale surface coatings, thin films and layers have at least one nanoscale dimension. In the following subsections, nanomaterials are described according to the primary (or core) chemical composition of nano-objects: carbon containing nanomaterials (e.g. fullerenes, carbon nanotubes); oxides nanomaterials (e.g. TiO_2 and ZnO); metal nanomaterials (e.g. Au); semiconductor nanomaterials (e.g. quantum dots); organic polymeric nanomaterials (e.g. dendrimers); and bio-inspired nanomaterials (e.g. capsid nanoparticles). Within these classes, different nanomaterials are listed in the order of decreasing necessary number of dimensions in nanoscale from 3D particles to fibers to layers.

3.1.1. Carbon containing nanomaterials

3.1.1.1. Fullerenes

Fullerenes are chemical entities which can be envisioned as spherical cages built from carbon atoms chemically bonded to three nearest neighbors. The best known example is a soccer-ball shaped C_{60} fullerene. Fullerene molecules can contain from 28 to more than 100 carbon atoms with some experimental studies reporting molecules containing up to 1 500 atoms with 8.2 nm diameter.[2] Existence of even larger fullerene molecules has been postulated from theoretical considerations.[3] Multi-shell fullerene-like nanoparticles referred to as carbon nano-onions, can range in size between 4 and 36 nm.[4] Fullerenes are actively investigated for a wide range of potential applications including: lithium-ion batteries, solar cells, fuel cells, oxygen and methane storage materials, additives to plastics, oil and rubber, and cancer and AIDS treatments.

3.1.1.2. Carbon black

Carbon black consists of partially amorphous material, organized into spherical or near-spherical particles fused together to give aggregates, weakly interacting to form agglomerates, usually further organized into macroscopic pellets.[5] Furnace black accounts for 98 % of the worldwide production and has an average aggregate diameter of 80-500 nm and an average primary particle size of 11-95 nm. The main industrial uses of carbon black are as a pigment and as reinforcing filler for rubber articles, in particular, tires.

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3.1.1.3. Carbon nanofibers

Carbon nanofibers (CNFs) are cylindrical or conical structures that have diameters ranging from a few to one hundred nanometers and lengths ranging from under micrometer to several millimeters. The internal structure is comprised of stacked curved graphite layers (or graphene sheets, see also section 3.1.1.5) that form cones (herringbone structure), cups (bamboo structure), rods (solid structure), or tubes (hollow structure).[6] The main distinguishing characteristic of nanofibers from nanotubes is the stacking of graphene sheets which make a non-zero angle with the fiber axis. When graphene sheets are parallel to the fiber axis, they form carbon nanotubes (see next section). Since there are “in-plane” and “interplane” components of transport and mechanical properties along the fiber axis, as well as presence of unsaturated bonds similar to graphite, carbon nanofiber characteristics differ from those of carbon nanotubes.

Carbon nanofibers are produced during chemical vapor deposition processes from carbon rich gases such as hydrocarbons over metal catalysts.[7] A greater control over carbon nanofiber structure and composition can be achieved with catalytic plasma-enhanced chemical vapor deposition.[8] Carbon nanofibers are produced on an industrial scale and find applications as polymer additives, gas storage materials and catalyst supports.[9]

3.1.1.4. Carbon nanotubes

Carbon nanotubes (CNTs) represent a diverse family of carbon-based materials based on a graphene sheet rolled up in the form of a tube. CNTs can be made up of one sheet (Single-Walled) or several sheets (Multi-Walled). Single-walled CNT can be open- or closed-ended depending on whether they are capped with fullerene halves at each end. Carbon nanotubes can have a diameter as small 0.4 nm and reach several centimeters in length.[10,11] Multi-walled form can reach 100 nm in diameter.[12]

Single-walled carbon nanotubes display metallic or semiconductive properties depending on how the graphene sheet is rolled up, and their electronic response can be tuned using elemental subsitution.[13] Carbon nanotubes have been predicted to be as much as sixty times stronger than steel and six times lighter.[14] They are considered excellent heat conductors, have a great capacity for molecular absorption and are chemically and thermally very stable.[15]

Applications which are currently being investigated include; polymer composites, electromagnetic shielding, electron field emitters, super capacitors, batteries, hydrogen storage and structural composites. Main synthesis methods for carbon nanotubes fall into two classes: those in which elemental carbon is vaporized typically by a laser or an electric arc and those in which the carbon is derived at lower temperature from a carbon source usually assisted by a catalyst or plasma.[16]

Commercial manufacturing and supply of carbon nanotubes at a large-scale production rate appears to be taking place in a number of countries.

3.1.1.5. Graphene nanosheet

Graphene sheet is a single layer of graphite structure which can be described as a hexagonal network of carbon atoms bonded to three nearest neighbors. Microscopic roughening through out-of-plane deformations makes graphene sheet effective thickness of about 1 nm. Graphene was shown to possess unique electronic, magnetic, optical and mechanic properties and might find applications in flat flexible electronic devices and coatings.[17] Micromechanical cleavage is presently the main method used to prepare this material.

3.1.2. Oxides

Metal oxide nanostructured materials in the form of agglomerated and aggregated nanoparticles are used mostly as paint and sunscreen additives and often coated to achieve desired properties. Main production methods are spray pyrolysis, laser ablation and solution phase synthesis.

Metal oxide nano-objects can be grown with a variety of simple shapes such as nanorod, nanotubes,[18] nanoflakes, and more complex structures such as nanobrushes, nanosprings, and nanobelts.[19] These nanostructures exhibit unique electronic properties and can find novel applications in optoelectronics, sensors, transducers, and medicines.

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Synthetic amorphous silica can be manufactured as a nanostructured material via gas-phase synthesis or wet chemical processes, such as precipitation or sol-gel process. The nanostructured material consists of primary particles within a range of 5-10 nm forming hard aggregates (1-40 µm). Primary particles do not exist as individual units; aggregation and agglomeration are predominant in particle formation and growth. Synthetic amorphous silica is currently used in a wide variety of industrial applications. Most of them are related to the reinforcement of various elastomers, the thickening of various liquid systems, the free-flow of powders or as a constituent of matting, absorbents and heat insulation material.[20,21]

3.1.3. Metals

Gold nanoparticles are one of the most extensively studied. Gold nanoparticles are characterized by a prominent optical resonance in the visible range, which is sensitive to environmental changes, size, and shape of the particles as well as to local optical interactions in resonant systems. This unique property of gold nanoparticles is utilized in a number of applications such as optical markers and as thermal targeted cancer treatment agent in medicine. Silver nanoparticles are produced in largest volumes among metal nanoparticles and used in numerous applications ranging from wound dressings to washing-machine disinfectant for its anti-microbial activity.[22]

Metal nanoparticles with well-defined size and shape can be synthesized using metal reduction from a solution phase.[23]

Metal nanowires such as cobalt, gold and copper-based can be conductive or semiconductive and could be used as interconnectors for the transport of electrons in nanoelectronic devices.[16] Nanowires are typically manufactured by involving a template followed by the deposition of a vapor to fill the template and grow the nanowire.[16] Deposition processes currently include Electrochemical Deposition and Chemical Vapor Deposition. The template might be formed by various processes including etching, or the use of other nanomaterials such as nanotubes.[16]

3.1.4. Quantum dots

Spherical nanocrystals from 1 to 10 nm in diameter composed of semiconductor materials often possess unique optical properties due to quantum effects, hence they are often called quantum dots. The number of atoms in quantum dots makes them neither an extended solid structure nor a molecular entity. The light emitted can be adjusted to the desired wavelength by changing the overall dimension.[24]

Quantum dots are used, among other purposes, as fluorescent probes in diagnostic medical imaging and in therapeutics; they are used for these purposes due to their optical properties and our ability to coat and modify their surfaces with peptides, antibodies, nucleic acids and other biologically important molecules.[25]

Currently, chemistry, physics and material science have provided methods for the production of quantum dots and are allowing tighter control on factors such as particle growth and size, solubility and emission properties. The most common method to produce quantum dots is by wet chemical colloidal processes.[16]

3.1.5. Organic polymeric nanomaterials

3.1.5.1. Dendrimers

Dendrimers are a new class of controlled-structure multi-branched polymers with nanoscale dimensions. They allow precise, atomic-level control of the synthesis of nanostructures according to the desired dimensions, shape and surface chemistry. They can display both hydrophilic and hydrophobic characteristics and can accommodate a wide variety of functional groups for medical applications. They are expected to be used in the medical and biomedical field.[26] Most syntheses of dendrimers involve the repetitious alternation of a growth reaction and an activation reaction such as the more traditional Michael reaction, or the Williamson ether synthesis, and more modern solid-phase synthesis, organo-metallic chemistry, organo-silicon chemistry, and organo-phosphorus chemistry.[26]

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3.1.5.2. Fibers

Nanofibers can be made of a wide variety of polymeric materials. The main manufacturing techniques are electrospinning and gas-blowing. These techniques allow for great flexibility in controlling chemical composition and physical parameters such as fiber diameter and length. Nanofiber scaffolds can be used in a number of applications such as sensors and ultrafiltration devices for liquid and gas phase.[27] Biodegradable polymer nanofibers can find numerous applications in medicine as scaffolds for tissue engineering, in controlled drug release, wound dressings, molecular separation, and bone restoration.[28]

3.1.6. Bio-inspired nanomaterials

Bio-inspired nanomaterials are generally materials in which a biological substance is trapped, encapsulated or adsorbed on the surface. They include a wide range of engineered assemblies of biological building blocks such as lipids, peptides and polysaccharides utilized as carriers for drugs, receptors, nucleic acids and imaging agents. Examples are polymeric micelles, protein cage architectures, viral-derived capsid nanoparticles, polyplexes, and liposomes[29] used in transport and optimal targeting of drugs. A number of formulations are under development for drug delivery via gastrointestinal and inhalation routes and skin applications.

Micelles are formed in solution as aggregates in which amphiphilic molecules are arranged in a spheroidal structure with hydrophobic cores shielded from the water by a mantle of hydrophilic groups. These dynamic systems, which are usually below 50 nm in diameter, are used for the systemic delivery of water-insoluble drugs. Drugs or contrast agents might be trapped physically within the hydrophobic cores or can be linked covalently to component molecules of the micelle.[29]

Liposomes are closed lipid bilayer vesicles that form by hydration of dry phospholipids. Drug molecules can be either entrapped in the aqueous space or intercalated into the lipid bilayer of liposomes, depending on the physicochemical characteristics of the drug. The liposome surface is amenable to modification with targeting ligands and polymers.[29]

Polyplexes are assemblies, which form spontaneously between nucleic acids and polycations or cationic liposomes (or polycations conjugated to targeting ligands or hydrophilic polymers), and are used in transfection protocols. The shape, size distribution, and transfection capability of these complexes depends on their composition and charge ratio of nucleic acid to that of cationic lipid/polymer. Examples of polycations that have been used in gene transfer/therapy protocols include poly-L-lysine, linear- and branched-poly(ethylenimine), poly(amidoamine), poly-ß-amino esters, and cationic cyclodextrin.[29]

Protein cage architectures and viral-derived capsid nanoparticles are formed by self-assembly of certain proteins.[29]

Building blocks of bio-inspired nanomaterials can be obtained from natural materials and using synthetic microbiology techniques,[30] while self-assembly often takes place in a liquid phase.

3.2. Production processes

3.2.1. Typical production processes

Methods typically used for the manufacturing of nanomaterials are:

- Aerosol generation such as flame pyrolysis, high temperature evaporation and plasma synthesis;
- Vapor deposition;
- Liquid phase methods: colloidal, self-assembly, sol-gel;
- Electropolymerization and electrodeposition;
- Electro-spinning for polymer nanofiber synthesis;
- Mechanical processes including grinding, milling and alloying.

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3.2.2. Aerosol generation methods

The aerosol generation method is used to produce a wide range of nanomaterials. This method is based on homogeneous nucleation of a supersaturated vapor and subsequent particle growth by condensation, coagulation and capture. The formation of vapor typically occurs within an aerosol reactor at elevated temperatures where often a super saturate of a solid is cooled into a background of gas. The methods used to produce nanomaterials are usually categorized by the heating or evaporation process and include:[16]

- Flame pyrolysis
- Furnace/hot wall reactors
- Laser induced pyrolysis

3.2.3. Vapor deposition methods

These methods are traditionally based on already well known and established methods for the manufacture of semiconductors. Here, vapor is formed in a reaction chamber by pyrolysis, reduction, oxidation and nitridation. The first step is the deposition of a few atoms. These first atoms form islands which spread and coalesce into a continuous film. Later, growth continues until thicker film develops.[16]

These methods have been used to produce nanofilms including TiO_2, ZnO and SiC.[16] Vapour deposition processes mediated by a catalyst are used to produce carbon nanotubes commercially.

3.2.4. Colloidal/self-assembly methods

The colloidal methods are also well established conventional wet chemistry precipitation processes in which solutions of different ions at required concentrations are mixed under controlled conditions of temperature and pressure which form insoluble precipitates.[16]

Recently, a rapidly expanding sub-set of colloidal methods called sonochemistry methods, where acoustic cavitation is used to control the process.[31] Here molecular precursors undergo chemical reactions because of the application of ultrasound radiation. It is the creation, growth and rapid collapse of a bubble that is formed in the liquid which is the main event. In this process, high temperatures and high cooling rates accompany the collapse of the bubble and nucleation centers formed whose growth is limited by the rapid collapse.[16]

Chalcogenides, metals and alloys including gold, cobalt and nickel as well as carbon and titania nanotubes have been produced using this method.[16]

3.2.5. Electrodeposition

Polymer nanofiber and metal nanowire films can be fabricated on an substrate through a controlled electropolymerization (polymers) or electrodeposition (metals) process.[32,33]

3.2.6. Electro-spinning

Electro-spinning method is a major method in the manufacture of polymer nanofibers. It utilizes electrical force to produce polymer fibers from polymer solutions or melts.[34]

3.2.7. Attrition methods

In attrition methods, size reduction is accomplished by grinding and milling and production of materials such as clay, coal and metals have been made.[16] Production rates in the order of tons per hour can be obtained using these methods.

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Bibliography

[1] Thess, A., Lee, R., Nikolaev, P., Dai, H.J., Petis, P., Robert, J., Xu, C.H., Lee, Y.H., Kim, S.G., Rinzler, A.G., Colbert, D.T., Scuseria, G.E., Tomanek, D., Fischer, J.E., and Smalley, R.E., Crystalline ropes of metallic carbon nanotubes, Science, 273, 483-487, 1996.

[2] Rietmeijer, F.J.M., Rotundi, A., Heymann, D. C-60 and giant fullerenes in soot condensed in vapours with variable C/H-2 ratio. Fullerenes, Nanotubes and Carbon Nanostructures 12 (3), 659-680, 2004.

[3] Siber, A. Shapes and energies of giant icosahedral fullerenes – Onset of ridge sharpening transition. European Physical Journal B 53 (3), 395-400, 2006.

[4] Sano, N., Wang, H., Alexandrou, I., Chhowalla, M., Teo, K.B.K., Amaratunga, G.A.J., Iimura, K. Properties of carbon onions produced by an arc discharge in water. Journal of Applied Physics 92 (5), 2783-2788, 2002.

[5] Ungar, T., Gubicza, J., Ribarik, G., and Pantea, C., Microstructure of carbon blacks determined by X-ray diffraction profile analysis, Carbon 40 (6), 929-937, 2002.

[6] Endo, M., Kim, Y. A., Hayashi, T., Fukai, Y., Oshida, K., Terrones, M., Yanagisawa, T., Higaki, S., and Dresselhaus, M. S., Structural characterization of cup-stacked-type nanofibers with an entirely hollow care. Appl. Phys. Lett. 80 (7), 1267-1269, 2002.

[7] Baker, R. T. K., Catalytic growth of carbon filaments, Carbon 27 (3), 315-323, 1989.

[8] Chen, Y., Wang, Z. L., Yin, J. S., Johnson, D. J., and Prince, R. H., Well-aligned graphitic nanofibers synthesized by plasma-assisted chemical vapor deposition, Chem. Phys. Lett. 272 (3-4), 178-182, 1997.

[9] De Jong, K. P., and Geus, J. W., Carbon nanofibers: Catalytic synthesis and applications, Cat. Rev. – Sci. Eng. 42 (4), 481-510, 2000.

[10] Zhu, H.W., Xu, C. L., Wu, D. H., Wei, B. Q., Vajtai, R., and Ajayan, P. M., Direct synthesis of long single-walled carbon nanotube strands, Science 296 (5569), 884-886, 2002.

[11] Wang, N., Tang, Z.K., Li, G.D., Chen, J.S., Single-walled 4 A carbon nanotube arrays, Nature, 408(6808), 50-51, 2000.

[12] Wang, H., Xu, Z., and Eres, G., Order in vertically aligned carbon nanotube arrays, Appl. Phys. Lett. 88, 213111, 2006.

[13] Burch, H. J., Davies, J. A., Brown, E., Hao, L., Contera, S. A., Grobert, N., Ryan, J. F., Electrical conductance and breakdown in individual CNx multiwalled nanotubes, Appl. Phys. Lett. 89, 143110, 2006.

[14] Demczyk, B.G., Wang, Y. M., Cumings, J., Hetman, M., Han, W., Zettle, A., and Ritchie, R. O., Direct mechanical measurement of the tensile strength and elastic modulus of multiwalled carbon nanotubes, Mater. Sci. Eng. A 334, 173-178, 2002.

[15] Harris, P. J. F., Carbon nanotubes and related structures. Cambridge University Press, 279 p, 1999.

[16] The Royal Society, Royal Academy of Engineering. Nanoscience and Nanotechnologies: Opportunities and Uncertainties. 2004. Available online at: http://www.nanotec.org.uk/finalReport.htm.

[17] Neto, A.C., Guinea, F., Peres, N.M. Drawing conclusions from graphene, Physics World 19 (11), 33-37, 2006.

[18] Lu, J. G., Chang, P., and Fan Z., Quasi-one-dimentional metal oxide materials- synthesis, properties and applications, Mat. Sci. Eng. R 52, 49-91, 2006.

[19] Wang, Z. L., Nanostructures of Zinc Oxide, Mat. Today June, 26-33, 2004.

© ISO 2008 – All rights reserved
Documents
Document Sharing Hub
Full document details: PD ISO/TR 12885:2008

[20] Ferch, H.,Toussaint, H.-E., Synthetic amorphous silicas in fine powder form: definitions, properties and manufacturing processes. Kautschuk Gummi Kunststoffe, 49, 589-596, 1996.

[21] ECETOC, Synthetic Amorphous Silica (CAS No. 7631-86-9), JACC No. 51, European Centre for Ecotoxicology and Toxicology of Chemicals, 2006.

[22] Panáček, A., Kvitek, L., Prucek, R., Kolář, M., Večeřová, R., Pizúrova, N., Sharma, V.K., Nevěčná, T., Zbořil, R., Silver Colloid Nanoparticles: Synthesis, Characterization, and Their Antibacterial Activity. J. Phys. Chem. B 110, 16248-16253, 2006.

[23] Cushing, B.L., Kolesnichenko, V. L., O'Connor, C. J. Recent Advances in the Liquid-Phase Synthesis of Inorganic Nanoparticles. Chem. Rev. 104, 3893-3946, 2004.

[24] Aitken, R. J., Creely, K. S., Tran, C. L., Nanoparticles: an occupational hygiene review. Sudbury, Suffolk, G.-B. HSE, 100p, 2004. http://www.hse.gov.uk/research/rrpdf/rr274.pdf.

[25] Smith A.,M., Gao, X., Nie, S., Quantum-Dot Nanocrystals for In-vivo Molecular and Cellular Imaging, Photochem. Photobiol. 80, 377-385, 2004.

[26] Tomalia, D. A., Birth of a new macromolecular architecture: dendrimers as quantized building blocks for nanoscale synthetic polymer chemistry, Prog. Polymer Sci. 30 (3-4), 294-324, 2005.

[27] Burger, C., Hsiao, B.S., Chu, B. Nanofibrous materials and their applications, Ann. Rev. Mat. Res. 36, 333-368, 2006.

[28] Zhang, Y., Lim, C.T., Ramakrishna, S., Huang, Z.M. Recent development of polymer nanofibers for biomedical and biotechnological applications. J. Mater. Sci. Mater. Med. 16 (10), 933-946, 2005.

[29] Moghimi, S.M., Hunter, A.C., Murray, J.C. Nanomedicine: current status and future prospects. FASEB Journal 19, 311-330, 2005.

[30] Asenjo, J.A., Bioreactor system design. Asenjo, J.A., Merchuk, J.C. Eds. Bioprocess technology; v. 21. Marcel Dekker, Inc.: New York, New York, 1994.

[31] Gedanken, A., Using sonochemistry for the fabrication of nanomaterials, Ultrasonics Sonochem, 11 (2), 47-55, 2004.

[32] Yu, X.F., Li, Y.X., Zhu, N.F., Yang, Q.B., Kalantar-zadeh, K., A polyaniline nanofibre electrode and its application in a self-powered photoelectrochromic cell, Nanotechnology 18, 1, 2007.

[33] Walter, E.C., Murray, B.J., Favier, F., Kaltenpoth, G., Grunze, M., Penner, R.M., Noble and coinage metal nanowires by electrochemical step edge decoration, J. Phys. Chem. B 106, 11407-11411, 2002.

[34] Huang, Z.M., Zhang, Y.Z., Kotaki, M., Ramakrishna, S., A review on polymer nanofibers by electrospinning and their applications in nanocomposites, Comp. Sci. Tech. 63, 2223-2253, 2003.

4. Hazard characterization

4.1. Health effects

The potential health risk of 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. Since nanotechnology is an emerging field, there are uncertainties as to whether the unique properties of engineered nanomaterials also pose unique occupational health risks. These uncertainties arise because of gaps in knowledge about the factors that are essential for evaluating health risks (e.g., routes of exposure, translocation of materials once they enter the body, and interaction of the materials with the body's biological systems). An important issue is whether the nanoscale version of a particular material poses risks that are significantly different in type or intensity than the macroscale forms of the same material.

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Results of existing studies in cell cultures (in vitro), animals (in vivo) or humans (epidemiological) on exposure and response to nanoscale or other respirable[1-4] particles, as well as available toxicity information about a given material in macroscopic form, provide a basis for preliminary estimates of the possible health effects from exposures to similar engineered materials on a nano-scale. However, it should be recognized that there are significant uncertainties and variables associated with predicting human health effects based on animal studies. Presently, in vitro cell culture methods are used mostly to delineate mechanisms of toxicity. In general, these in vitro data can not be extrapolated to humans without additional information (e.g. in vivo data). Initial experimental studies in animals have shown that the biological response (whether beneficial or detrimental) to certain incidental or engineered nanoparticles can be greater than that of the same mass of larger particles of similar chemical composition.[5-12] In addition to particle number and combined surface area, other particle characteristics might influence the biological response, including solubility, shape, charge and surface chemistry, catalytic properties, adsorbed pollutants (e.g. heavy metals or endotoxins), as well as degree of agglomeration.[13-15]

Often nanoparticle surfaces are intentionally modified with coatings or functionalized in order to prevent agglomeration of particles and to achieve desired properties, e.g. pharmacological activity. Such modifications, as well as the contamination of particle surfaces with impurities can lead to changes in biological responses. More research is underway to study the influence of particle properties on interactions with living organisms and the potential for adverse effects.

4.1.1. Basic principles and uncertainties

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, it is known from studies in humans that a greater proportion of inhaled nanoparticles will deposit in the alveolar region of the respiratory tract (both at rest and with exercise) compared to larger particles.[16,17] However, it has to be realized that nanoparticles might agglomerate and that these agglomerates can deposit in other areas of the respiratory tract or possibly cannot be inhaled at all. Further, animal studies indicate that nanoparticles after initial exposure can be translocated to other organs in the body, although it is not well known how this might be influenced by the chemical and physical properties of the nanoparticles.[18-22] Additional uncertainties are introduced by the difficulties in predicting human health effects based on animal studies. There might also be the potential for greater dermal and gastro-intestinal uptake of nanoparticles when compared to larger particles. Evidence from nanotoxicological studies (in vitro and animal studies) suggests that exposure to some nanoparticles might have the potential to cause cell/ tissue/ systemic toxicity. Due to their small size, nanoparticles have the potential to cross cell membranes and interact with subcellular structures, such as mitochondria and the nucleus (and some nanoparticles have been shown to cause oxidative damage and impair some function of cells in culture).[23,24] Animal studies have indicated that some nanoparticles are more biologically active due to their greater surface area per mass compared with larger-sized particles of the same chemistry when dose response relationships are expressed as mass.[5-12] The greater surface area per mass of nanoparticles compared to larger particles is a fundamental contributor to the greater chemical reactivity and utility of nanoparticles for industrial, commercial, and medical applications, but it also raises concern about the potential for adverse health effects in workers exposed to nanoparticles.

4.1.2. Potential relevance of health effects information about incidental or naturally-occurring nanoparticles and nanofibers

While there is limited information on the health effects of engineered nanoparticles, there is a larger body of research on the health effects of incidental nanoparticles (e.g. diesel exhaust particulate[25,26] and welding fumes[27]). The biological mechanisms of particle-related lung responses (e.g., oxidative stress, inflammation, and production of cytokines, chemokines, and cell growth factors) appear to be a consistent lung response to incidental respirable, including nanoscale, particles[13,28,29] (contaminants such as transition metals[30] might also contribute to the lung response[31]). Although the composition and thus the physicochemical characteristics of incidental and engineered nanoparticles can differ substantially, the toxicological and dosimetric principles derived from studies of incidental nanoparticles might be relevant to assessing the health effects of engineered particles.

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There is a very large body of research on the health effects of certain respirable fibers. For example, it is well established that particular forms of asbestos are causative factors in otherwise rare, occupationally-derived, malignant mesotheliomas[32] and other lung disorders (including pulmonary interstitial fibrosis, pleural plaques, calcification and thickening).[33] The harmful effects of fibers are driven by three important factors: length, diameter and persistence.[34-37] It is difficult to draw conclusions regarding the health effects of engineered nanoscale fibers based on asbestos studies, but they suggest that particle properties of size, shape and composition are important factors influencing the toxicity of nanoparticles. There have been shorter-term studies of the effects of single-walled carbon nanotubes[38,39] and multi-walled carbon nanotubes[40] in the lungs of rats and mice (administered by intratracheal instillation or pharyngeal aspiration). These studies have shown unusual inflammatory and fibrogenic reactions in the lungs, including transient inflammation followed by early onset of fibrosis at mass doses lower than those causing fibrosis from quartz or carbon black.[38,39]

4.1.3. Relationship between toxicity and surface area, surface chemistry, and particle number

To the extent that nanoparticles might pose increased hazards, the most significant factors might relate to the greater number or surface area of nanoparticles compared with that for the same mass concentration of larger particles. This hypothesis is based primarily on the pulmonary effects observed in studies of rodents exposed to various types of poorly soluble nanostructured materials in the form of agglomerated and aggregated nanoparticles or larger respirable particles (e.g., titanium dioxide, carbon black, barium sulfate, carbon black, diesel soot, coal fly ash, and toner). These studies found that for a given mass of particles, poorly soluble nanostructured materials in the form of agglomerated and aggregated nanoparticles produced greater observable effect than larger particles of similar chemical composition and surface properties. Dose-response relationships obtained in animal studies for poorly-soluble and low toxicity particles appear consistent across particle sizes when dose is expressed as particle surface area.[5-12,41-45] The mechanisms by which these materials exhibit higher levels of toxicity at smaller particle sizes (on a mass basis) appear to involve pulmonary inflammation, oxidative stress, and tissue injury.[13,28,29] The biological activity of particles is affected by their number and their physical and chemical properties, including size, surface area, solubility, shape, crystal structure, charge, catalytic activity and chemistry.[5-9,13,14,46-49]

Through engineering, the properties of nanomaterials can be modified. For example, recent in vitro studies have shown that the in vitro cytotoxicity of fullerenes (measured as cell death) and carbon nanotubes can be reduced by several orders of magnitude by modifying the surface chemistry of the fullerene molecules and carbon nanotubes (e.g., by hydroxylation).[50,51] Cytotoxicity studies in vitro with quantum dots have shown that the type of surface coating can have a significant effect on cell motility and viability.[52-54]

4.1.4. Inflammatory response to nanoparticles

A variety of nanoparticles ranging from carbon-based combustion products to transition metals, having entered tissues and cells, can elicit generalized inflammatory and acute phase responses which incorporate the release of signaling molecules such as chemokines, cytokines, C-reactive protein and fibrinogen (a coagulant).[55-61] In addition, macrophage and neutrophil activation is also associated with the production of reactive oxygen species. Macrophages are the well-known surveillance cells in the tissues which react with particulate matters, including nanoparticles to produce these biological responses. Exposure to different nanoparticles has been found to modulate, in different ways, the defence/inflammatory capacities of macrophages.[62] In response to cytokines and chemokines, plasma proteins and neutrophils migrate from the blood to start inflammation. One of the roles of neutrophils in the inflammation is to destroy foreign bodies by proteolysis as well as reactive oxygen species. However, the excess or prolonged defence reactions against foreign bodies by these cells damage the tissues, too. When the process of macrophage ingestion and (attempted) proteolytic breakdown go awry or when nanoscale particles become internalised in non-immune-related cell types (e.g. parenchymal cells), intracellular molecular defenses are initiated which result in the new expression of protective genes. When these protective mechanisms (e.g., antioxidants) are depleted, tissue injury and disease might occur.[29,63] Several broad intracellular protective responses are known, from cell culture and animal studies, to be induced by the exposure to particulate/fibrous materials (carbon black, carbon nanotubes, ambient air particles, and nanostructured titania): the pro-inflammatory cytokines and relevant genes; the antioxidant-response element-inducible genes and proteins; and the stress (or “heat shock”) response proteins.[55-61] Consequently, effects such as persistent inflammatory responses and gene inductions are likely to represent precursors of downstream pathological conditions. It is important to note that contaminants, such as metal catalysts or bacterial endotoxins (lipopolysaccharides), contributed to the

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induction of inflammatory responses observed in experimental toxicological studies of nanoparticles. Specifically, the unpurified single-walled carbon nanotubes which contained more than 20 % by weight of iron induced stronger pulmonary inflammation than purified counterparts;[30] and the conventional formulations of gold nanoparticles which contained significant amounts of endotoxins stimulated immune cells *in vitro,* and the improvement of formulation process, which diminished the endotoxin contamination, also effectively reduced the bilogical responses.[64]

4.1.5. Animal and cell-culture studies

4.1.5.1. Carbon containing nanomaterials

Major manufactured nanoscale carbon containing materials are carbon nanotubes (CNTs), carbon nanofibers, fullerenes and carbon black. Single-walled CNT can be described as a single sheet of graphite rolled to form a seamless cylinder (see also Chapter 3.1.1). This new class of material offers excellent electrical, mechanical and thermal properties. Due to such unique advantageous material properties an increased production and use of CNTs makes it likely that human exposure will occur.

A number of toxicological studies of CNTs have been performed in recent years (see references cited in [15], [65], [66]). These studies have suggested that the biological response to CNTs in cell-culture and animal studies can vary widely depending on the test material and the test method applied. The nature of the test material depends on the method of production and post-production treatment resulting in different levels of impurities (metals, organic molecules, other forms of carbon, support material etc.) and different structures (atomic structure, number of walls, agglomeration state, etc.) and geometries (diameter, length, deformations) of carbon nanotubes. Therefore it is essential that certain physical characteristics of a test material are determined and reported in conjunction with any investigation of their hazards.[67]

Biomarkers of oxidative stress were elevated in an *in vitro* assay after incubation with single-walled carbon nanotubes.[30] Single-walled CNTs purified by acid treatment have been shown to produce adverse effects, including acute inflammation with early onset, yet progressive, fibrosis and granulomas in the lungs of mice after a single pharyngeal aspiration. Application of carbon black at identical mass doses did not induce granulomas or alveolar wall thickening and pulmonary inflammation, and damage was less pronounced.[38] Although these results, obtained by pharyngeal aspiration, suggest some hazardous potential of single-walled CNTs by inhalation, the actual human inhalation toxicity of single-walled CNTs has not been determined yet.

Purified[ii]ground and intact multi-walled carbon nanotubes administered intratracheally to rats were found to be biopersistent (still present in the lung after 60 days) and induced inflammatory and fibrotic reactions.[40] Pulmonary lesions induced by CNTs characterized by the formation of collagen-rich granulomas were caused by the accumulation of large CNT agglomerates in the airways. Ground CNTs were better dispersed in the lung parenchyma and also induced inflammatory and fibrotic responses. Chrysotile asbestos and carbon black were included as reference materials. As expected, asbestos induced inflammatory and fibrotic reactions while carbon black only showed inflammatory reaction. [40]

There have also been studies on the effects of exposure to carbon nanotubes on the skin and eyes. An *in vitro* study reported that unpurified single-walled carbon nanotubes caused a significant decrease in cellular viability and biomarkers of oxidative stress with a dose-response relationship, as well as a significant increase in lipid peroxides on human epidermal keratinocytes.[68] This study concluded that dermal exposure to unrefined single-walled carbon nanotubes might lead to dermal toxicity in exposed workers. Another *in vitro* study using human fibroblasts and keratinocytes indicates that single-walled CNTs, functionalized with peptides, are capable of penetrating the cell membrane.[69] However, the application of a filter saturated with a solution containing fullerene soot with a high content of single-walled CNTs during a patch test did not cause irritation or allergies in volunteers.[70] Ocular instillation of an aqueous suspension of nanotubes in a modified Draize test with rabbits did not cause irritation.[70]

There have been a few toxicity studies of functionalized carbon nanotubes. Numerous effects have been reported and the toxicity seems to vary greatly with the nature of the functional groups. An *in vitro* study with

[ii] "Purified" in this context means treated with acid to remove metal contaminants.

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lung tumor cells showed that the toxicity of unpurified multi-walled carbon nanotubes and nanostructured carbon black obtained by grinding of graphite increases after their surfaces are chemically functionalized with carbonyl, carboxyl and hydroxyl groups.[71] On the other hand, another *in vitro* study using cultured human dermal fibroblasts showed that cytotoxicity of purified single-walled carbon nanotubes functionalized with phenyl-SO_3H and phenyl-$(COOH)_2$, decreases as the degree of functionalization increases.[51] In vivo mice studies showed that water-soluble, single-walled CNTs functionalized with diethylenetriaminepentaacetate and labeled with indium (^{111}In) for imaging purposes, were not retained in either liver or spleen and were rapidly cleared from systemic blood circulation through the renal excretion route after intravenous administration.[72]

4.1.5.2. Oxides

Experimental studies in animals have shown that at equivalent mass doses, poorly soluble nanostructured metal oxides in the form of agglomerated or aggregated nanoparticles (e.g., titanium dioxide, aluminum oxide, and manganese dioxide) are more potent than larger particles of similar composition, in causing pulmonary inflammation, tissue damage, and lung tumors in animals.[5-9,41,43,44] For these and other poorly soluble particles, a consistent dose-response relationship is observed when dose is expressed as particle surface area.[6,7,13,42,45] These animal studies suggest that for nanostructured materials and larger particles with similar chemical properties, the toxicity of a given mass dose will increase with decreasing particle size due to the increasing surface area. In addition to particle size and surface area, other physical and chemical properties of particles are known to influence toxicity, including solubility, shape, surface reactive sites, charge, and crystal structure.[13,14,47,48,73] For poorly soluble particles of relatively low toxicity, some animal studies have identified doses that were not associated with observed adverse responses. For example, a recent animal study reported mass doses of either fine or nanostructured TiO_2 in rats at which the lung responses did not significantly differ from controls, while crystalline silica caused more severe lung responses at the same mass dose.[74,75]

4.1.5.3. Metals

In vitro studies indicate that some metal nanoparticles can exhibit acute inflammatory effects in animals related to the ability of metal ions to generate reactive oxygen species. The dose-response relationships differ for different metals, which might relate to the physico-chemical properties and mechanisms of toxicity. For example, recent studies using rat liver derived cell lines *in vitro* indicate that silver nanoparticles (15 and 100 nm) at 5 - 50 µg/ml showed a significant cytotoxicity to the cells, whereas other particles such as aluminum nanoparticles (30 and 103 nm) and micrometer scale tungsten particles (27 µm), had no measurable effects at the same mass doses.[76] *In vitro* studies of mouse spermatogonia cell lines reported cytotoxic effects of silver nanoparticles (15 nm).[77] Peters *et al.* studying the behaviour and viability of human endothelial cells *in vitro*, observed that cobalt and nickel nanoparticles were incorporated into the vacuoles of the cells.[78] Cobalt nanoparticles were found to cause inflammation and to be cytotoxic, while nickel nanoparticles did not produce these effects. In contrast, an *in vivo* study found nickel nanoparticles was more toxic than cobalt nanoparticles, and that the toxicity correlated with the free radical activity.[79] In other *in vivo* studies, nickel nanoparticles was found to be more toxic to the lungs than the same mass dose of micrometer scale nickel particles,[80] and cobalt nanoparticles were more toxic than micrometer scale cobalt particles.[81] In an oral gavage study, copper nanoparticles caused severe toxicity and injury to the kidney, liver, and spleen, while microscale copper particles did not.[82] Colloidal gold nanoparticles, which have been developed for therapeutic and diagnostic uses, did not generate any toxicity in mice by an intravenous injection at a therapeutic dose level.[83]

4.1.5.4. Semiconductors

The main application of semiconductor nanoparticles is as quantum dots. Health hazard properties of quantum dots depend on a host of factors arising from their chemical structure and environmental conditions. Size, charge, concentration, bioactivity of the surface coating, and oxidative, photolytic, and mechanical stability contribute to their toxicity.[84] The long-term stability of the complexes and their complete degradation before elimination needs to be further evaluated given that some of their constituents, such as Pb, As, Cd, and Tl, are potentially highly toxic.[85] Coatings can inhibit quantum dot degradation which might result in the release of toxic constituents, as well as loss of quantum dot fluorescence in vivo.[86]
In vitro studies suggest that some quantum dots can be cytotoxic, with dose-response relationship. For example, CdTe quantum dots caused rat pheochromocytoma cell death *in vitro* indicated by chromatin

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condensation and membrane blebbing.[87] Smaller positively charged quantum dots were more toxic than larger, equally charged, quantum dots with the former localized in the nuclear compartment and the latter in the cytosol. Authors explained this by the presence of Cd^{2+} ions, free radical formation, or interaction with intracellular components leading to loss of function. Other, *in vitro* studies, reported that CdSe quantum dots were cytotoxic to liver cells and that surface oxidation of the quantum dots produced Cd^{2+} ions, recognized as carcinogenic.[52,88] Encapsulation of the quantum dots with ZnS tended to reduce this effect and it declined to almost zero with encapsulation by bovine serum albumin. The *in vitro* cytotoxicities of realgar (As_2S_2, a semiconducting material which is also used in some traditional medical formulations) with small sizes (100 - 150 nm) to human umbilical vein endothelial cells were stronger than those of larger particles (200 - 500 nm).[89] The surface area-dependent cytotoxicity of realgar particles can be explained by the amount of released active ingredients in the incubation medium from the particles, which might be highly dependent on the amount of surface area.

Some in vivo studies showed no observed ill effects: mice injected with amphiphilic polyacrylic acid polymer-coated quantum dots and with polyethyleneglycol amine conjugated quantum dots;[90] mice injected with CdSe/ZnS quantum dots.[91]

4.1.5.5. Organic polymeric nanomaterials

Certain incidental organic nanoparticles have been shown to pose acute toxic hazard upon human inhalation exposure. Among nanoscale particles, freshly-generated polytetrafluoroethylene (PTFE) fume (generated at temperatures of more than 425 ^{o}C) is known to be highly toxic to the lungs. Freshly-generated PTFE fume (15 nm) caused hemorrhagic pulmonary edema and death in rats exposed to less than 60 µg/m^3.[46] In contrast, aged PTFE fume was much less toxic and did not result in mortality, which was attributed to changes in surface chemistry as well as increase in particle size from accumulation (>100 nm).[13,92] While PTFE fume differs from engineered nanoparticles, these studies illustrate properties of incidental nanoparticles that have been associated with an acute toxic hazard.

Toxicity of nanoscale dendrimers is related to the nature of monomers and dendrimer synthesis.[93] Specifically, *in vitro* and *in vivo* animal studies showed that nanoscale dendrimers with positively charged surface groups similar to other biological macromolecules can destabilize cell membranes and cause cell lysis.[94] Thus, biological response to nanoscale dendrimers can be tailored through functionalization of the surface dendrimer sites. Nature of the dendrimer core also is also thought to affect its biological activity. For example, it has been suggested that dendrimers with aromatic interior can cause hemolysis through hydrophobic membrane contact.[95] Higher generation dendrimers (larger in size) have been found to be more cytotoxic.[96]

4.1.5.6. Bio-inspired nanomaterials

Engineered bio-inspired nanomaterials can potentially produce the full range of health responses observed with naturally occurring nanomaterials, from the benign and beneficial (such as insulin and growth hormone) to adverse and even lethal (such as protein biotoxins “designed” by nature to be toxic).

It was shown that delivery of biologically active bio-inspired nanoparticles into the blood circulation through oral administration can be facilitated by bile acids and proteinase inhibitors.[97] Recently, transdermal delivery of intact, biologically active protein medications, such as insulin, has been shown to be possible in the presence of phage peptide chaperones.[98] It appears that the mechanism of penetration is not specific to insulin and involves interactions between the phage peptide and the skin, facilitating a transfollicular route of insulin transport through the skin. These drug applications could potentially result in accidental exposures of workers during drug production and administration in health care settings.

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4.1.6. Observations from epidemiological studies involving fine and nanoscale particles

Initial epidemiological studies in workers exposed to aerosols, including incidental fine and nanoscale particles, have reported lung function decrements, adverse respiratory symptoms, chronic obstructive pulmonary disease, and fibrosis.[2-4,27,99,100] In addition, some studies have found elevated lung cancer among workers exposed to certain incidental nanoscale particles, e.g., diesel exhaust particulate[25,26] and welding fumes.[27] 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.[101,102] The implications of these studies for engineered nanoparticles, which might 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.[103-106] Some epidemiological studies have shown adverse health effects associated with exposure to the incidental nanoscale particulate fraction of air pollution,[107-111] although uncertainty exists about the role of incidental nanoscale 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 nanoscale carbon particles, changes were observed in the expression of adhesion molecules by blood leukocytes, which might relate to possible cardiovascular effects of incidental nanoscale particle exposure.[112] Controlled clinical studies in the laboratory have shown deposition of incidental nanoscale dusts throughout the pulmonary tree, accompanied by cardiovascular problems.[13,113-115]

Human studies of nanoparticles are primarily based on people exposed to fumes (e.g., welding), diesel exhaust or particulate ambient air pollution. Epidemiological studies aimed specifically at engineered nanoscale particles have not yet been performed, with the exception of TiO_2[12] and carbon black.[2-4] Therefore, the extrapolation of the findings associated with air pollution and particulates generally (which might include incidental nanoscale particles) to engineered nanoparticles is uncertain. However, the potential risks from exposure to nanomaterials, including occupational exposure, should be considered.

4.2. Physical hazards

4.2.1. Fire (exothermic events)

Although insufficient information exists to predict the potential fire and explosion risk associated with nanoscale powders, nanoscale combustible material might present a higher risk than coarser material of similar quantity due to exothermic events arising from catalytic reactions or due to a lowering of minimum ignition temperatures.[116] Decreasing the particle size of combustible materials can reduce minimum ignition energy and increase combustion rate, leading to the possibility of relatively inert materials becoming combustible. Dispersions of combustible nanomaterials in air might present a greater safety risk than dispersions of micro- and macro-materials with similar compositions. Dust explosiveness, minimum ignition energy and ignition temperature are typical means of characterizing safety relevant aspects of dusts.

Nanoparticles and nanostructured porous materials have been used for many years as effective catalysts for increasing the rate of reactions or decreasing the necessary temperature for reactions to occur in liquids and gases. Depending on their composition and structure, some nanomaterials might initiate catalytic reactions and increase their fire and explosion potential that would not otherwise be anticipated from their chemical composition alone.[117]

Additionally, some nanomaterials are designed to generate heat through the progression of reactions at the nanoscale. Such materials might present a fire hazard that is unique to engineered nanomaterials. The greater activity of nanoscale materials forms a basis for research into nanoenergetics. For instance, nanoscale Al/MoO_3 thermites ignite more than 300 times faster than corresponding micrometer-scale material.[118]

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4.2.2. Safety considerations in manufacturing nanomaterials

The manufacture of nanomaterials, as with any other novel materials, can be either on a small pilot scale, including research and development activities, or full scale. Presently, nanomaterial manufacturing can include several high-energy processes, such as flame pyrolysis, laser induced pyrolysis, laser vaporization, thermal plasma, microwave plasma, sputtering, and laser ablation, which present specific safety issues. Typical safety hazards in these processes include handling of high pressure cylinders, low pressure apparatus, inert and toxic gases, high temperature objects, operation of high electrical currents, electro-magnetic radiation emitting devices, and high intensity light sources (including ultra-violet, infra-red, and visible light) and lasers. Working under such hazardous conditions requires implementing appropriate work practices or following respective laboratory safety guidelines.

Bibliography

[1] ISO 7708:1995, *Air quality — Particle size fraction definitions for health-related sampling*.

[2] Büchte, S. F., Morfeld, P., Wellmann, J., Bolm-Audorff, U., McCunney, R. J., and Piekarski, C., Lung cancer mortality and carbon black exposure : a nested case-control study at a German carbon black production plant, J. Occup Environ. Med. 48 (12),1242-1252, 2006.

[3] Morfeld, P., Büchte, S. F., Wellmann, J., McCunney, R. J., and Piekarski, C., Lung cancer mortality and carbon black exposure: Cox regression analysis of a cohort from a German carbon black production plant, J. Occup Environ. Med. 48 (12), 1230-1241, 2006.

[4] Morfeld, P., Büchte, S. F., McCunney, R. J., and Piekarski, C., Lung cancer mortality and carbon black exposure: uncertainties of SMR analyses in a cohort study at a German carbon black production plant, J. Occup Environ. Med. 48 (12),1253-1264, 2006.

[5] Oberdörster, G., Ferin, J., Gelein, R., Soderholm, S. C., and Finkelstein, J., Role of the alveolar macrophage in lung injury—studies with ultrafine particles, Environ. Health Perspect. 97,193–199, 1992.

[6] Oberdörster, G., Ferin, J., Lehnert, B. E., Correlation between particle-size, in-vivo particle persistence, and lung injury, Environ. Health Perspect. 102 (S5), 173–179, 1994.

[7] Oberdörster, G., Ferin, J., Soderholm, S., Gelein, R., Cox, C., Baggs, R., and Morrow, P. E., Increased pulmonary toxicity of inhaled ultrafine particles: due to lung overload alone? Ann. Occup. Hyg. 38 (Suppl. 1), 295-302, 1994.

[8] Lison, D., Lardot, C., Huaux, F., Zanetti, G., and Fubini, B., Influence of particle surface area on the toxicity of insoluble manganese dioxide dusts, Arch. Toxicol. 71 (12), 725-729, 1997.

[9] Tran, C. L., Buchanan, D., Cullen, R. T., Searl, A., Jones, A. D., and Donaldson, K., Inhalation of poorly soluble particles. II. Influence of particle surface area on inflammation and clearance, Inhal. Toxicol. 12 (12), 1113–1126, 2000.

[10] Brown, D. M., Wilson, M. R., MacNee, W., Stone, V., Donaldson, K., Size-dependent proinflammatory effects of ultrafine polystyrene particles: A role for surface area and oxidative stress in the enhanced activity of ultrafines, Tox. Applied Pharm. 175 (3), 191-199, 2001.

[11] Bermudez, E., Mangum, J. B., Wong, B. A., Asgharian, B., Hext, P. M., Warheit, D. B., and Everitt, J. I., Pulmonary responses of mice, rats, and hamsters to subchronic inhalation of ultrafine titanium dioxide particles, Tox. Sci. 77, 347-357, 2004.

[12] Hext, P.M., Tomenson, J.A., and Thompson, P., Titanium dioxide: inhalation toxicology and epidemiology, Ann. Occ. Hyg. 49 (6), 461-472, 2005.

[13] Oberdörster, G., Oberdörster, E., and Oberdörster, J., Nanotoxicology: an emerging discipline evolving from studies of ultrafine particles, Environ. Health Perspect. 113 (7), 823–839, 2005.

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Full document details: PD ISO/TR 12885:2008

[14] Maynard, A. M., Kuempel, E. D., Airborne nanostructured particles and occupational health, J. Nanopart. Res. 7(6), 587-614, 2005.

[15] Donaldson, K., Aitken, R., Tran, L., Stone, V., Duffin, R., Forrest, G., and Alexander, A., Carbon Nanotubes: a Review of Their Properties in Relation to Pulmonary Toxicology and Workplace Safety, Toxicol. Sci. 92 (1), 5-22, 2006.

[16] ICRP, Human respiratory tract model for radiological protection. Oxford, England: Pergamon, Elsevier Science Ltd., International Commission on Radiological Protection Publication No. 66, 1994.

[17] Kim, C. S., Jaques, P. A., Analysis of total respiratory deposition of inhaled ultrafine particles in adult subjects at various breathing patterns, Aerosol Sci. Technol. 38, 525-540, 2004.

[18] Takenaka, S., Karg, D., Roth, C., Schulz, H., Ziesenis, A., Heinzmann, U., Chramel, P., and Heyder, J., Pulmonary and systemic distribution of inhaled ultrafine silver particles in rats, Environ. Health Perspect. 109 (suppl. 4), 547-461, 2001.

[19] Kreyling, W. G., Semmler, M., Erbe, F., Mayer, P., Takenaka, S., Schulz, H., Oberdorster, G., and Ziesenis, A., Translocation of ultrafine insoluble iridium particles from lung epithelium to extrapulmonary organs is size dependent but very low, J. Toxicol. Environ. Health 65 (20), 1513–1530, 2002.

[20] Oberdörster, G., Sharp, Z., Atudorei, V., Elder, A., Gelein, R., Lunts, A., Kreyling, W., and Cox, C., Extrapulmonary translocation of ultrafine carbon particles following whole-body inhalation exposure of rats, J. Toxicol. Environ. Health 65 Part A(20), 1531–1543, 2002.

[21] Oberdörster, G., Sharp, Z., Atudorei, V., Elder, A., Gelein, R., Kreyling, W., and Cox, C., Translocation of inhaled ultrafine particles to the brain, Inhal. Toxicol. 16 (6–7), 437–445, 2004.

[22] Semmier, M., Seitz, J., Erbe, F., Mayer, P., Heyder, J., Oberdorster, G., and Kreyling, W. G., Long-term clearance kinetics of inhaled ultrafine insoluble iridium particles from the rat lung, including transient translocation into secondary organs. Inhal. Toxicol. 16 (6-7), 453-459, 2004.

[23] Geiser, M., Rothen-Rutishauser, B., Kapp, N., Schurch, S., Kreyling, W., Schulz, H., Semmler, M., Im Hof, V., Heyder, J., and Gehr, P., Ultrafine particles cross cellular membranes by nonphagocytic mechanisms in lungs and in cultured cells, Environ. Health Perspect. 113 (11), 1465-1560, 2005.

[24] Moller, W., Brown, D. M., Kreyling, W. G., and Stone, V., Ultrafine particles cause cytoskeletal dysfunctions in macrophages: role of intracellular calcium, Part. Fibre Toxicol. 2 (7), 1-12, 2005.

[25] Garshick, E., Laden, F., Hart, J. E., Rosner, B., Smith, T. J., Dockery, D. W., and Speizer, F. E., Lung cancer in railroad workers exposed to diesel exhaust, Environ. Health Perspect. 112 (15), 1539–1543, 2004.

[26] Steenland, K., Deddens, J., and Stayner, L., Diesel exhaust and lung cancer in the trucking industry: exposure-response analyses and risk assessment, Am. J. Ind. Med. 34 (3), 220–228, 1998.

[27] Antonini, J. M., Health effects of welding. Crit. Rev. Toxicol. 33 (1), 61–103, 2003.

[28] Donaldson, K., Tran, L., Jimenez, L. A., Duffin, R., Newby, D. E., Mills, N., MacNee, W., and Stone, V., Combustion-derived nanoparticles: a review of their toxicology following inhalation exposure, Part. Fibre Toxicol. 2 (10), 2005.

[29] Knaapen, A. M., Borm, P. J., Albrecht, C., and Schins, R. P., Inhaled particles and lung cancer. Part A: Mechanisms, Int. J. Cancer 109 (6), 799-809, 2004.

[30] Kagan, V. E., Tyurina, Y. Y., Tyurin, V. A., Konduru, N. V., Potapovich, A. I., Osipov, A. N., Kisin, E. R., Schwegler-Berry, D., Mercer, R., Castranova, V., and Shvedova, A. A., Direct and indirect effects of single walled carbon nanotubes on RAW 264.7 macrophages: role of iron, Toxicol. Lett. 165, 88–100, 2006.

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Full document details: PD ISO/TR 12885:2008

[31] McNeilly, J. D., Jimenez, L. A., Clay, M. F., MacNee, W., Howe, A., Heal, M. R., Beverland, I. J., Donaldson, K., Soluble transition metals in welding fumes cause inflammation via activation of NF-kappaB and AP-1, Toxicol. Lett. 158 (2), 152-157, 2005.

[32] Robison, B. W., Lake, R. A., Advances in malignant mesothelioma, N. Eng. J. Med. 353 (15), 1591-1603, 2005.

[33] U. S. ATSDR; Agency for Toxic Substances and Disease Registry, Toxicological Profile for Asbestos, 2001.

[34] Stanton, M. F., and Wrench, C., Mechanisms of mesothelioma induction with asbestos and fibrous glass, J. Natl. Cancer Institute 48, 797-821, 1972.

[35] Stanton, M. F., Laynard, M., Tegeris, A., Miller, E., May, M., and Kent, E., Carcinogenicity of fibrous glass: pleural response in the rat in relation to fiber dimension, J. Natl. Cancer Institute, 58, 587-603, 1977.

[36] Suzuki, Y., Yuen, S.R., and Ashley, R., Short, thin asbestos fibers contribute to the development of human malignant mesothelioma: pathological evidence. Int. J. Hyg. Env. Health, 208 (3), 201-210, 2005.

[37] U. S. NIOSH, Asbestos Bibliography (Revised). DHHS (NIOSH) Publication Number 1997-162, 1997. Available on line at www.cdc.gov/niosh/97-162.html.

[38] Shvedova, A. A., Kisin, E. R., Mercer, R., Murray, A. R., Johnson, V. J., Potapovich, A. I., Tyurina, Y. Y., Gorelik, O., Arepalli, S., Schwegler-Berry, D., Unusual inflammatory and fibrogenic pulmonary responses to single walled carbon nanotubes in mice, Am. J. Phys. Lung Cell. Mol. Phys. 289 (5), L698-L708, 2005.

[39] Lam, C. W., James, J. T., McCluskey, R., and Hunter, R. L., Pulmonary toxicity of single-wall carbon nanotubes in mice 7 and 90 days after intratracheal instillation, Toxicol. Sci. 77, 126–134, 2004.

[40] Muller, J., Huaux, F., Moreau, N., Misson, P., Heilier, J.-F., Delos, M., Arras, M., Fonseca, A., Nagy, J. B., and Lison, D., Respiratory toxicity of multi-wall carbon nanotubes, Toxicol. Appl. Pharmacol. 207, 221-231, 2005.

[41] Lee, K. P., Trochimowicz, H. J., and Reinhardt, C. F., Pulmonary response of rats exposed to titanium dioxide (TiO2) by inhalation for two years, Toxicol. Appl. Pharmacol. 79, 179–192, 1985.

[42] Oberdörster, G., Yu., The carcinogenic potential of inhaled diesel exhaust: a particle effect? J. Aerosol Sci. 21, S397–S401, 1990.

[43] Oberdörster, G., Ferin, J., Finkelstein, G., Wade, P., and Corson, N., 1990. Increased pulmonary toxicity of ultrafine particles? II. Lung lavage studies, J. Aerosol Sci. 21, 384-391, 1990.

[44] Heinrich, U., Fuhst, R., Rittinghausen, S., Creutzenberg, O., Bellmann, B., Koch, W., and Levsen, K., Chronic inhalation exposure of wistar rats and 2 different strains of mice to diesel-engine exhaust, carbon-black, and titanium-dioxide, Inhal. Toxicol. 7 (4), 533–466, 1995.

[45] Driscoll, K. E., Role of inflammation in the development of rat lung tumors in response to chronic particle exposure, in Particle overload in the rat lung and lung cancer: implications for human risk assessment, Mauderly, J. L., McCunney, R. J. Taylor & Francis, Philadelphia, PA, 1996, pp.139–152.

[46] Oberdörster, G., Gelein, R. M., Ferin, J., and Weiss, B., Association of particulate air pollution and acute mortality: involvement of ultrafine particles? Inhal. Toxicol. 7 (1), 111-124, 1995.

[47] Duffin, R., Tran, C. L., Clouter, A., Brown, D. M., MacNee, W., Stone, V., and Donaldson, K., The importance of surface area and specific reactivity in the acute pulmonary inflammatory response to particles, Ann. Occup. Hyg. 46, 242-245, 2002.

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Full document details: PD ISO/TR 12885:2008

[48] Brunner, T. J., Wick, P., Manser, P., Spohn, P., Grass, R. N., Limbach, L. K., Bruinink, A., and Stark, W. J., In vitro cytotoxicity of oxide nanoparticles: comparison of asbestos, silica, and the effect of particle solubility, Environ. Sci. Technol. 40, 4374-4381, 2006.

[49] Limbach, L. K., Wick, P., Manser, P., Grass, R. N., Bruinink, A., and Stark, W. J., Exposure of Engineered Nanoparticles to Human Lung Epithelial Cells: Influence of Chemical Composition and Catalytic Activity on Oxidative Stress, Environ. Sci. Technol. 41 (11), 4158-4163, 2007.

[50] Sayes, C. M., Fortner, J. D., Guo, W., Lyon, D., Boyd, A. M., Ausman, K. D., Tao, Y. J., Sitharaman, B., Wilson, L. J., Hughes, J. B., West, J. L., and Colvin, V. L., The differential cytotoxicity of water-soluble fullerenes, Nano Lett. 4 (10), 1881-1887, 2004.

[51] Sayes, C. M., Liang, F., Hudson, J. L., Mendez, J., Guo, W., Beach, J. M., Moore, V. C., Doyle, C. D., West, J. L., Billups, W. E., Ausman, K. D., Colvin, V. L., Functionalization density dependence of single-walled carbon nanotubes cytotoxicity in vitro, Toxicol. Lett. 161 (2), 135-42, 2006.

[52] Shiohara, A., Hoshino, A., Hanaki, K., Suzuki, K., and Yamamoto, K., On the cytotoxicity of quantum dots, Microbiol. Immunol. 48 (9), 669-675, 2004.

[53] Lovric, J., Cho, S. J., Winnik, F. M., Maysinger, D., Unmodified cadmium telluride quantum dots induce reactive oxygen species formaition leading to multiple organelle damage and cell death, Chemistry & Biology 12 (11), 1127-1234, 2005.

[54] Hoshino, A., Fujioka, K., Oku, T., Suga, M., Ssaki, Y., Ohta, T., Physicochemical properties and cellular toxicity of nanocrystal quantum dots depend on their surface modification, Nano Lett. 4 (11), 2163-2169, 2004.

[55] Barlow, P. G., Clouter-Baker, A. C., Donaldson, K., MacCallum, J., and Stone, V., Carbon black nanoparticles induce type II epithelial cells to release chemotaxins for alveolar macrophages. Part. Fiber Toxicol. 2, 1-14, 2005.

[56] Brown, D. M., Donaldson, K., Borm, P. J., Schins, R. P., Dehnhart, M., Gilmour, P., Jimenez, L. A., and Stone, V., Calcium and ROS-mediated activation of transcription factors and TNF-alpha cytokine gene expression in macrophages exposed to ultrafine particles. Am. J. Physiol. Lung Cell. Mol. Physiol. 286, L344-L353, 2004.

[57] Cui, D., Tian, F., Ozkan, C. S., Wang, M., and Gao, H. Effect of single wall carbon nanotubes on human HEK293 cells, Toxicol. Lett. 146, 73-85, 2005.

[58] Hetland, R. B., Cassee, F. R., Refsnes, M., Schwarze, P. E., Låg, M., Boere, A. J., and Dybing, E., Release of inflammatory cytokines, cell toxicity and apoptosis in epithelial lung cells after exposure to ambient air particles of different size fractions, Toxicol. In Vitro 18, 203-212, 2004.

[59] Ramage, L., Guy, K., Expression of C-reactive protein and heat-shock protein-70 in the lung epithelial cell line A549, in response to PM10 exposure. Inhal. Toxicol. 16, 447-452, 2004.

[60] Stearns, R. C., Paulauskis, J. D., Godleski, J. J., Endocytosis of ultrafine particles by A549 cells, Am. J. Respir. Cell. Mol. Biol. 24, 108-115, 2001.

[61] Stone, V., Tuinman, M., Vamvakopoulos, J. E., Shaw, J., Brown, D., Petterson, S., Faux, S. P., Borm, P., MacNee, W., Michaelangeli, F., and Donaldson, K., Increased calcium influx in a monocytic cell line on exposure to ultrafine carbon black, Eur. Respir. J. 15, 297-303, 2000.

[62] Lucarelli, M., Gatti, A. M., Savarino, G., Quattroni, P., Martinelli, L., Monari, E., Boraschi, D., Innate defence functions of macrophages can be biased by nano-sized ceramic and metallic particles, Eur. Cytokine Network, 15 (4), 339-346, 2004.

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