



# **Joint NNI-ChI CBAN and SRC CWG5 Nanotechnology Research Needs Recommendations**

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## Summary

- The National Nanotechnology Initiative (NNI) encouraged the formation of consultative working groups to help define the research needs in emerging nanotechnology from the industry perspective.
- The chemical industry formed the NNI-Chemical Industry Consultative Board for Advancing Nanotechnology (NNI-ChI CBAN) with a subgroup working on nanotechnology ESH issues.
- The semiconductor industry formed the SRC NNI Consultative Working Group # 5 (CWG5) focused on nanotechnology ESH issues
- The chemical industry and the semiconductor industry found that they have many of the same concerns in nanotechnology ESH.
- The two working groups began meeting jointly and developed the following research needs document
- The document has 5 main areas:
  - Metrics for nanoscale particle toxicity
  - Exposure monitoring methodologies
  - Risk assessment methodology
  - Testing strategy for toxicity
  - Societal communication and education

## **Developing the best metric for nanoscale particle toxicity**

### Problem / Issue /Need:

Dose is typically measured as the mass of a substance to which a person is exposed over some time period and is usually expressed as milligram (mass) per kilogram (a measure of body weight) per day) or as milligram (mass) per m<sup>3</sup> (volume of air) per hour. For nanoscale materials, these standard metrics may not be sufficient or the most relevant parameters for measuring the potential exposures to nanoscale materials. Because the number and surface area of nanoscale materials may be very high relative to mass, alternative metrics are likely to be more important. Although dimensional metrology may provide alternative dose “metrics”, other characterization, additional characterization may also be relevant.

- Dose metrics
  - (mg/kg)
  - size of primary particle, the degree or proportion of aggregates, and agglomerates; aerodynamic size; number concentration; surface area
- Chemical composition (identity, impurities; surface characteristics)
- Shape and structure
- Dissolution / durability characteristics
- Presence, nature, purity, effects, stability, influence of additives, shells and coatings

### Current State of Knowledge:

Ferin et al. (1992) studied ultrafine and fine TiO<sub>2</sub> particles, concluding that the translocation of particles into the interstitium appeared to be a function of the number of particles, and the process appeared to be related to the particle size, the delivered dose, and the delivered dose rate. The relationship of particle surface area to inflammogenicity, with some limitations in using this metric for all aspects of toxicity, was discussed by Oberdorster (1996). The potential importance of aggregate size (of small primary particles) and stability are useful areas of investigation. Information from past studies on carbon black and titanium dioxide may be of value as these have been studied quite extensively before the current interest and focus on nanoscale primary particles.

G. Oberdorster (Univ. Rochester) leads a Department of Defense funded research project (Air Force Multi-disciplinary University Research Initiative) to identify the relationship between physicochemical characteristics and toxicological properties of nanoscale materials. Number and surface area were noted as likely to be critical dosimetrics. Other physicochemical characteristics such as size, surface area and porosity, crystallinity and chemical composition are to be assessed with respect to their role as effect modifiers. Results expected from the

DOD study will guide the selection of "dose" for nanoscale particle/material exposure assessment.

An additional challenge is that some of the above metrics (including particle size) will be specific to exposure media. Size of particles in bulk samples may not be the same as the size in the delivery vehicle. If exposure relevant studies are conducted which address the metrics noted above, it may be possible to assess whether some or all of these are critical determinants of toxicity. Studies which use simplified assessment or exposure procedures (e.g. *in vitro* studies, or *intra tracheal* administration) will not necessarily accurately reflect particle / particle or particle / exposure media interactions, nor the normal line of physiological defenses. In toxicology, delivered dose (at target site) is more accurate, particularly when comparing responses between species.

#### Evaluation of tools and capabilities:

##### *A Composition*

Conventional methods for the accurate determination of the chemical composition of materials may need to be modified or enhanced in order to be applied to nanoscale materials. Chemical characterization at or near the atomic level will likely be required for the accurate assessment of the chemical composition of nanoscale particles. Applicable techniques may include secondary ion mass spectrometry, X-ray photoelectron spectroscopy, Auger-electron spectroscopy, aberration-corrected analytical electron microscopy, X-ray microanalysis, scanning probe microscopies (SPM) including AFM and STM, and near-field optical methods. Novel methods for the determination of the core atomic composition as well as ligand composition and surface reactivity such as single particle mass spectrometry may also be applicable. Surface charge of nanoscale particles may be determined by using zeta potential measurement techniques though standard operating procedures are lacking. The presence and the nature of additives should be addressed.

##### *B Dimensional Metrology*

Indirect methods which may be applicable to the characterization of the size and number distribution of nanoscale engineered particle populations include: differential light scattering, analytical ultra centrifugation, ion mobility classification, and small angle scattering using X-ray or neutron sources. In addition there is progress in the development of automated electron microscopic methods (direct method) for the rapid analysis and screening of a large number of nanoscale engineered particles.

Direct methods which may be applicable to size characterization of individual or small numbers of nanoscale particles or materials containing engineered nanoscale particles include: electron microscopies, (SEM & TEM), scanning

tunneling microscopy, atomic force microscopy, and super-resolution optical microscopy.

Separation techniques, such as liquid chromatography, size exclusion chromatography, and capillary electrophoresis may be applicable to the determination of the size-distribution of particles associated with nanoscale engineered materials.

Improved measurement methods for particles that are less than 5 nm are needed. Correlations studies of the various methods for determining particle size such as electron microscopy, differential light scattering, and analytical ultra centrifugation need to be carried out to determine what if any differences there are in their dimensional metrologies.

### *C Surface Area*

The surface area as well as surface charges of engineered nanoscale particle populations is believed to be a critical parameter in toxicity. Surface area measurements for nanotechnology have been reported from at least three different methods. All of these methods employ assumptions which do not necessarily apply to nanoscale particles.

1. The method developed by Brunauer, Emmet, and Teller or BET which is a classical method based on the adsorption and release of nitrogen or other gasses. The application of BET to nanoscale particle measurements is not validated and often the assumption is made that the primary particles are all spherical, with constant diameter and density.
2. Diffusion-charging methods in which an electrometer is used to determine the amount of charge imparted to the particle population. The magnitude of the charge is related back to particle size and surface area. These instruments are available commercially but have not been validated.
3. Epiphaniometry in which the alpha activity is measured from the adsorption of Pb 211 onto particle surfaces. This method has not been validated.

### *D Shape and Structure*

The determination of the shape and structure of nanoscale particles associated with nano scale engineered materials will likely require characterization at or near the atomic-scale. The morphology of nano surfaces are also very relevant. Methods which may be applicable to this area include aberration-corrected transmission electron microscopy, scanning tunneling microscopy, and atomic force microscopy. Currently none of these methods are applicable to the analysis of large numbers of particles or large amounts of materials. Aspect ratios and chirality of nanoscale engineered particles may be determined by electron and optical microscopy, gas chromatography mass spectrometry, and liquid

chromatography mass spectrometry though modifications or enhancements are likely necessary to apply these methods to nanoscale engineered materials. The degree of nanoscale particle agglomeration may be determined by ion mobility classification and zeta potential although these methods have not been validated on nanoscale particles.

Recommendations:

Develop a set of metrics to accurately characterize the toxicology of nanoscale materials. Nanoscale materials share the characteristic of having at least one dimension in the 1 to 100 nm range; however they are not a single class of chemicals with common chemical, physical, and biological properties. Different nanoscale particles or nanoscale materials can vary in their toxicological properties, and, therefore, it is unlikely that there will be a one-size-fits-all answer to the question of metrics. A taxonomy should be developed to facilitate and improve comparative research efforts

In toxicology, delivered dose (in bloodstream or at target site) is a more accurate determinant of potential toxicity, particularly when comparing responses between species or extrapolating the results found in one species to predict the effects in another species. As our knowledge of nanoscale materials and toxicology develops, delivered dose may be a worthwhile research metric. Currently, however, delivered dose is difficult to measure and is not a routine metric for toxicology or workplace evaluations.

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## **Develop Exposure Monitoring Methodologies for Nanoscale Particles**

### Problem / Issue / Need:

There is limited information available on the mechanisms of nanoparticle toxicity. Due to the lack of toxicology information, there is uncertainty as to the appropriate exposure measurement methodology for human exposures to nanoparticles. It is critical that exposure measurement accurately reflect exposure health risk as they will be utilized to validate exposure controls and monitor workplace exposure levels. Therefore exposure monitoring methodologies to quantitatively assess both occupational and non occupational exposures to nanoscale particles that may be produced during their manufacturing or use is needed. There is also a need for standardized sampling and analytic techniques to quantify inhalation, oral and dermal exposure to nanoscale particles.

### Current State of Knowledge:

Commonly used analytical techniques designed to quantify exposures to chemical substances do not take into account potential differences in toxicity of nanoscale particles or the fact that nanoscale particles can behave differently than particles larger than 100 nanometers or conventional molecules. There are currently no standardized sampling and analytical techniques to quantify exposures to unbound nanoscale particles. Current research indicates that mass and bulk chemistry may be less important than particle size, surface area, and surface chemistry (or activity) for nanostructured materials [Oberdörster et al. 1992, 1994]. Research is still ongoing into the relative importance of these different exposure metrics, and how to best characterize exposures against them.

Current state of the art analytical techniques rely on laboratory instrumentation that measure parameters or characteristics of nanoscale particles such as; surface area, surface characteristics, particle count, particle mass, particle size distribution, etc. Currently, there is uncertainty within the scientific community as to what parameter or characteristic should be measured to quantify exposure. and which measured parameter will be most effective in determining or predicting toxic effects from nanoscale particle exposures.

### Recommendations:

Given the need to quantify human exposures to nanoscale particles in both occupational and non-occupational settings we recommend the development of a standardized nanoparticle exposure monitoring techniques and analytical methodologies. Key factors to be consider should include the determination of appropriate exposure measurement techniques that can be used to quantify oral, dermal and inhalation exposures to nanoparticles. The selected exposure measurement techniques should reflect the best indicator of harmful exposure potential based on known toxicological properties of nanoparticles.

References:

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<http://www.particleandfibretoxicology.com/content/2/1/8>

## **Development of a Risk Assessment Methodology for Exposure to Nanoscale Particles**

### Problem / Issue /Need:

There is a need to develop a standardized Risk Assessment Methodology to assess both occupational and non occupational health risk from potential exposures to nanoscale particles.

### Current State of Knowledge:

Occupational health risks associated with manufacturing and using nanoscale materials are not yet clearly understood. The rapid growth of nanotechnology is leading to the development of new materials, devices and processes that lie far beyond our current understanding of environmental and human impact. Many nanoscale materials and devices are formed from nanometer-scale particles (nanoparticles) that are initially produced as aerosols or colloidal suspensions. Exposure to these materials during manufacturing and use may occur through inhalation, dermal contact and ingestion. Currently minimal information is available on dominant exposure routes, potential exposure levels and material toxicity. What information does exist comes primarily from the study of ultrafine particles (typically defined as particles smaller than 100 nanometers).

Studies have indicated that low solubility ultrafine particles are more toxic than larger particles on a mass for mass basis. There are strong indications that particle surface area and surface chemistry are primarily responsible for observed responses in cell cultures and animals. There are also indications that ultrafine particles can penetrate through the skin, or translocate from the respiratory system to other organs. Research is continuing to understand how these unique modes of biological interaction may lead to specific health effects.

Workers within nanotechnology-related industries have the potential to be exposed to uniquely engineered materials with novel sizes, shapes and physical and chemical properties, at levels far exceeding ambient concentrations. To understand the impact of these exposures on health, and how best to devise appropriate exposure monitoring and control strategies, much research is still needed. Until a clearer picture emerges, the limited evidence available would suggest caution when potential exposures to nanoparticles may occur

There are currently no published methodologies on conducting qualitative risk assessment for potential exposures to nanoscale particles. Current state of the art risk analysis techniques are ineffective when applied to nanoscale particles due to the unknown toxicity and potential unknown exposure routes of unbound nanoscale particles. Specifically, there is limited toxicity information available on nanoscale particles or specific chemical compositions of nanoscale particles. Also there is limited information regarding the aerosol characteristics of nanoscale particles or their behavior in airstreams and on exposure mechanisms and the degree of health hazard posed by different exposure routes.

Recommendations:

Given the need for an appropriate risk assessment methodology for potential exposures to nanoscale particles in both occupational and commercial product use applications, we recommend the development of a nanoscale particle exposure assessment methodology. Key factors to be considered should include the ability to determine potential risk from handling/use of nanoscale particles based on conditions of use and potential toxicity of nanoscale particles. Risk assessment models should be developed to enable exposure risk assessment on nanoscale particles with unknown or limited toxicity information. The Risk assessment models should develop a framework for evaluating hazards and predicting risk of exposure to nanoscale materials

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## Develop a Testing Strategy for Assessing the Toxicity of Nanoscale Particles

### Problem / Issue /Need:

There is a need for an approach to evaluating nanoscale materials for mammalian and environmental toxicology. The established series of tests (e.g. EPA OPPTS Harmonized Guidelines and OECD Guidelines for the Testing of Chemicals) is a valuable basis for evaluating nanoscale materials. These tests are designed to evaluate a wide range of toxicological / biological responses. Also, there is flexibility in how some of these tests are applied. For example, to study the inhalation toxicology for aerosols, a holding period (satellite group) is sometimes used to assess potential changes in effects over time. This may be appropriate in some cases when insoluble materials are being evaluated. There are specific concerns relating to nanoscale material assessment that should be addressed. For example the traditional dose metric (weight / body weight) may not be sufficient due to the large surface area to mass of nanoscale materials. Characterizing test materials is another area that may have nanoscale material-specific requirements. Consideration should be given to exposure-relevant testing. For many applications, exposure to NM or free nanoscale particulates may not occur. If there is feasible exposure, the surrounding matrix (gas, liquid or solid) should be considered, as well as the potential importance of exposure route (inhalation, oral, dermal).

### Current State of Knowledge:

Recent reviews, including Oberdorster et al. (2005), Hoet et al. (2004), and the Royal Society (2004) cover broad aspects of nanoscale materials and nanotechnology. Although there have been toxicological evaluations of nanoscale materials such as carbon black and titanium dioxide, and some hazard evaluations of nanoscale materials; there are limited numbers of broad toxicological safety assessments on newly developed nanoscale materials. Simplified hazard identification tests have been conducted by non-physiologic administration (e.g. intra tracheal administration, *in vitro* tests). Examples include Lam et al., 2004 and Warheit et al., 2004. The bolus delivery of the test material over a few seconds *during intra tracheal* instillation can overwhelm normal lung defenses and cause effects that do not occur at physiologically relevant lower doses or lower dose rates predicted from inhalation exposure. *In vitro* assays can be a useful tool for examining the mechanisms of action of the different nanoscale materials, but there are significant limitations of *in vitro* results in hazard identification, i.e., the intricate pharmacokinetics that occurs in the body but absent in cell cultures. Another limitation of the currently available *in vitro* data is the high concentrations of nanoscale materials commonly used in the *in vitro* studies, which can be orders of magnitude higher than the predicted absorbed dose from environmental or occupational exposure. These have helped to demonstrate potential effects, but a more relevant exposure evaluation is needed to determine the relevance for human risk assessment.

Evaluation of tools and capabilities:

The methods described by EPA and OECD are largely applicable. Specific challenges include selecting relevant test materials, characterizing the test materials, administering the materials in a relevant manner (oral, dermal, or inhalation), measuring and characterizing the test material in the delivery matrix, measuring the test material in the biological matrix (blood, excreta, etc.) and any specific enhancements to the standard tests (perhaps satellite groups, pharmacokinetic assessment). Although the harmonized OECD approach to testing should form a basis for a comprehensive evaluation, improvements in understanding the potential effects of nanoscale materials may also come from more mechanistically oriented evaluations and specialized approaches to reported effects of ultrafines, such as cardiovascular toxicity.

Recommendations:

Given the number of nanoscale materials currently in research and development and the number of nanoscale materials finding their way to consumer products, we recommend the development of a screening/prioritizing strategy for the hazard identification of nanoscale materials with the highest EHS concerns. Key factors to be considered in the development of a screening/prioritizing strategy include the production volume of the nanoscale material, potential for occupational, consumer, and environmental exposure, physio-chemical properties indicative of potential toxicity, validation of available in vitro and short-term toxicity assays for the testing of nanoscale materials.

Consideration should be given to exposure-relevant testing, as results from exposure-relevant studies will provide the most useful information for assessing the potential risks from exposure to nanoscale materials. For many applications, exposure to NM or free nanoscale particulates may not occur. If there is feasible exposure, the surrounding matrix (gas, liquid or solid) should be considered, as well as the potential importance of exposure route (inhalation, oral, dermal). Determine what nanoscale and nanotechnology related materials could be most usefully evaluated. This might include materials that have some information already available and could serve as reference points (silica, carbon black, titanium dioxide), and newly created materials (carbon nanotubes, C-60 fullerenes) with incomplete toxicological information.

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## **EHS/Societal Research Communication/Education Needs related to Nanotechnology**

### Problem/Issue/Need:

As an emerging technology with potential environmental, health, safety and societal impacts, it is of critical importance that research results, both positive and negative, are communicated publicly in an open and credible manner. It should be noted that such communications should not be limited to EHS information, but also include messages that include new applications, societal impacts and benefits of the new technology. The public perception of a new technology can be significantly helped or hindered dependent on the communication mechanisms employed early in the development of the technology. Nanotechnology finds itself in the early stages of development when the general public is beginning to form opinions with regards to its benefits and/or detriments. A clear, coordinated, credible and international communication strategy is needed.

### Current State of Knowledge:

Nanotechnology research breakthroughs and promises of new applications for nanotechnology appear in the press on a regular basis. Likewise, research identifying potential concerns with the new technology is beginning to surface and be covered by the mainstream media. To date, it appears that most Americans hold a generally positive view toward nanotechnology. A July 2004 survey funded by the National Science Foundation (NSF) and conducted by North Carolina State University (NCSU) found that:

- Approximately 70% were “somewhat” or “very” hopeful about nanotechnology
- 80% were not worried at all about the science

However, more than 80% of those polled claimed to know “little” or “nothing” about nanotechnology and could not correctly respond to factual questions about the new technology. In addition, 60% of those surveyed claimed “not much trust” that business would minimize the risks of the technology. This sentiment suggests that strong public support may not be deeply rooted and that it will be critical for the government to take the lead on communications to the public.

Another survey (September/October 2004) by the University of Wisconsin-Madison Survey Center (UWSC) produced similar results regarding knowledge of nanotechnology to those of the NCSU findings, but raised additional questions. The U. Wisconsin researchers found that knowledge of nanotechnology was not a good predictor of attitudes toward nanotechnology. While those more knowledgeable survey respondents today have more favorable attitudes toward nanotechnology, the researchers concluded that this was because they are more trusting of scientists overall and are getting their information from the largely positive coverage in science media. The authors of the survey conclude that while knowledge of nanotechnology will likely increase in the future, the public's

perception will be largely shaped by the mass media (non-technical) as opposed to scientifically-based reporting. They further state that this may result in negative messages and shifts in perception along the line of the “Frankenfood” labels for genetically modified foods, and thus more negative attitudes toward nanotechnology.

Most recently, a study by the Project on Emerging Nanotechnologies, supported by the Woodrow Wilson International Center for Scholars and the Pew Charitable Trusts, confirmed many of the findings by NCSU and UMSC, but brought forth some interesting new findings. The study was completed with a focus group of 177 individuals and confirmed that the public is very interested in the potential for new technologies in areas such as medical applications, new consumer products, environmental protection, and nutrition, among others. However, the respondents expressed skepticism that industry and government would adequately address the potential risks associated with the technology. 55% felt that government oversight beyond voluntary standards is needed to manage EHS risks. More than 70% called for more pre-launch testing, more product information and more proof that workers and the environment were protected. Finally, participants consistently called for more information and to have a voice in the process.

In Europe, the EHS and societal concerns regarding nanotechnology appear to be more visible to the general public. For example, in the UK, Cambridge University and Greenpeace will kick-off a 5 week “NanoJury” in September to publicly weigh the pros and cons of nanotechnology and have a group of 20 “jurors” deliver a verdict on the technology. Although somewhat sensational, such open dialogues and education of the public may prove beneficial for technology acceptance, particularly in geographies which may be more skeptical of the science.

Recently, the EU has launched a new website to communicate the findings for two EHS-related nanotechnology projects funded by the European Commission’s Sixth Framework program – Impart-Nanotox. Likewise, several US agencies (including EPA, NSF and NIOSH) have established websites to communicate their EHS-related research and funding activities. In addition, NSF is expected to announce in upcoming months the creation of a research center to examine the societal implications of nanotechnology. Such steps represent excellent progress toward improving the nanotechnology communication to the public at large.

#### Recommendations:

In order to effectively manage the communication of nanotechnology research, advancements and EHS-related concerns, it is recommended that the following actions be taken:

(1) Coordinated communication for EHS impacts, societal impacts and overall benefits of nanotechnologies.

- Establish website(s) for communication targeted at the average citizen (non-technical).
- Provide summary of significant research findings (both positive and negative – risks and benefits) in a non-technical manner so that the general public can understand the implications.
- Develop communication collateral targeted at the general public – pamphlets, fact sheets, handouts, FAQs that present a balanced view of the issues.
- Create publicly available, searchable database of research papers, relevant studies and findings. (ICON has established a preliminary database)
- Establish conference(s) to publicly review research progress on EHS related issues and invite public input and participation.
- Create groups of stakeholders from industry, government, academia and NGOs to work together on communication strategies. One example of such a group is the International Council on Nanotechnology (ICON).

(2) Coordinated communication of EHS and societal research findings across major geographies including US, Europe and Asia.

- Form cross-geography communication mechanisms and information sharing on EHS related research findings – OECD may be a potential mechanism for accomplishing this goal.
- Sponsor international conference to share research findings and encourage dialogue between geographies.

(3) Ongoing survey/monitoring of public opinion in the US, Europe and Asia.

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NCSU Survey Press Release:

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NNI Environmental, Health and Safety Website:

<http://www.nano.gov/html/society/EHS.htm>

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