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Nanomaterials: The Good, the Bad, and the Ugly

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Introduction

The Nanotechnology Revolution is projected to be in full swing by 2015, creating more than $1 trillion in global commerce.[1] Whether engineered nanomaterials are produced or used in a laboratory setting, pilot plant or industry, the understanding of what nanomaterials are, their uses and the potential environmental, health, and safety hazards are essential to an environmental, health, and safety (EH&S) professional. The purpose of this case study is to present how one academic medical center approaches the use of engineered nanomaterials in research. From formation of a committee, identification of safety champions, staff education to create buy-in, guideline development, and hazard communication, this presentation embraces the "Good, Bad, and Ugly" of safe nanomaterial research.

Wake Forest University Baptist Medical Center (WFUBMC) is an integrated healthcare system that operates 1,187 acute care, rehabilitation and long-term care beds, outpatient services, and community health and information centers. The Medical Center's component institutions carry out a joint mission of patient care, education, research, and community service. The partnership includes three major members: Wake Forest University School of Medicine (WFUSM) and Wake Forest University Physicians, both part of Wake Forest University Health Sciences, and North Carolina Baptist Hospital. WFUBMC has 11,763 employees, of which 4,602 are Wake Forest University School of Medicine employees. The Medical Center operates a total of 5,778,108 square feet of building space, of which 1,602,912 square feet are designated as School of Medicine space. In total, the School of Medicine has sixty-seven buildings spread out over three campuses, including 215,000 square feet of animal facilities and 47,000 research animals, covering seventeen species.

One nanometer is one billionth of a meter. The "nanoscale" ranges from 100 nm down to the size of atoms (about 0.2 nm). At this scale, the properties of engineered nanomaterial are different from that of the macro-material with the same chemical composition. Nanomaterials have larger surface area when compared to an equal mass of the same material in macro form. At the nanoscale, chemicals are more reactive, and potentially more toxic. Strength and electrical properties are affected, and changes occur in the optical and the magnetic behavior of the materials. There are many different types of nanomaterials, carbon nanotubes (CNTs) (single wall (SWNT) and multi-wall (MWNT)), carbon black, fullerenes, nanoclays (silicon dioxide and titanium dioxide), polymeric, metals such as silver and gold nanoparticles, and quantum dots.

The research staff at WFUSM procures nanomaterials in a number of ways. They may purchase nanomaterials from the Center for Nanotechnology and Molecular Materials at Wake Forest University, which manufactures single-wall nanotubes (SWNT), multi-wall nanotubes (MWNT) and fullerenes. Researchers may also opt to synthesize their own nanomaterials in their labs, obtain them from a collaborator at another university, or purchase nanomaterials from various vendors. The type of nanomaterials currently being used at WFUSM are carbon nanotubes (CNTs) such as SWNT, DWNT, MWNT; carbon nanotubes with metals; fullerenes; silver and gold nanoparticles; and quantum dots.

The Good

The good news about the Nanotechnology Revolution is that nanomaterial uses are truly broad in scope with myriad applications both occurring and envisioned. Many industries stand to benefit, from industrial to consumer goods to biomedicine. Molecular switches, solar cells, composites, and semiconductors are a few of the applications for industrial use. For consumer use, these materials can be found in appliances, food and beverages, textiles, filtration, sports equipment, electronics, and cosmetics. Biomedical applications include nanomaterials utilized as drug carriers, tumor imaging, cell-targeted therapy, cell sensors and microchips, cell and tissue scaffolds, and wound dressings. Research involving the various types of nanomaterials at WFUSM seeks to find applications in the field of biomedicine and bioengineering.

In industries, employees who produce or develop nanomaterials are protected by health and safety regulations. Like industry, many of these regulations apply to universities and academic teaching facilities. Some of the common Occupational Safety and Health Administration (OSHA) regulations that would cover nanomaterial research are the general duty clause, hazard communication, and personal protective equipment (PPE). A less common OSHA regulation in industry, but pertinent to universities and teaching facilities, is the Occupational Exposure to Hazardous Chemicals in Laboratories regulation. Environmental Protection Agency (EPA) regulations pertaining to hazardous waste, air emissions and effluent discharge may affect nanomaterial research. The U.S. Department of Agriculture’s Animal Welfare Information Center (AWIC) and the Department of Health and Human Services Office of Laboratory Animal Welfare (OLAW) regulate animal research.

A variety of committees are chartered by the University to address chemical, biological, radiological and animal uses as required by the various agencies. Wake Forest University School of Medicine (WFUSM) has a Chemical Safety Committee, an Institutional Biosafety Committee, a Radiation Committee, and an Institutional Animal Care and Use Committee (IACUC).

The Occupational Exposure to Hazardous Chemicals in Laboratories regulation (1910.1450) requires the development of a chemical hygiene plan for laboratories and, if necessary, to form a chemical hygiene committee. At WFUSM, the Environmental Health & Safety Department (EH&S) has developed a general chemical hygiene plan for all laboratories. A Chemical Safety Committee has been formed, with rotating faculty members, and meets quarterly to review safety and health issues, along with pertinent chemical research protocols. Research protocols are required for chemicals that are considered particularly hazardous (i.e., carcinogens, reproductive toxins, highly acute toxicity, select agent toxins, hazardous drugs, air or water reactive and nanomaterials).

OSHA has established Permissible Exposure Limits (PELs) for many chemicals to protect employees from overexposure. However, no exposure standards have been established for nanomaterials. Some of the chemicals used to manufacture nanomaterials are listed in Table 1. These PELs were established without consideration for how these substances may interact with biological systems at the nanoscale.

Table 1. Permissible Exposure Limits

SUBSTANCE	PEL
Aluminum oxide	10 mg/m^3
Carbon black	3.5 mg/m^3
Magnesium oxide	10 mg/m^3
Silver, metal	0.1 mg/m^3
Iron oxide	5 mg/m^3
Silica, crystalline	0.25 mg/m^3
Chromium, metal	0.5 mg/m^3
Copper, dusts	1 ng/m^3
Titanium dioxide	10 mg/m^3
Tin, metal	2 ng/m^3

Historically, PELs, various federal, state, and local regulations, toxicological data, and proven engineering controls, administrative controls, and personal protective equipment have done an adequate job of protecting laboratory workers at WFUSM. However, nanomaterials present a dilemma.

The Bad

The bad news about the Nanotechnology Revolution is that the properties that make nanomaterials such an exciting and fast-evolving field of use and research are the same properties that create many unknowns for the EH&S professional. In the fall of 2008, the Chemical Safety Committee received its initial set of Chemical Safety Protocols for nanomaterials. This was the first time the subject of nanomaterial research had been brought forth for committee review. EH&S realized that the current protocol form did not do an adequate job of asking the principal investigator to delineate the hazards associated with nanomaterial research. Many Chemical Safety Committee members, including EH&S personnel, lacked the expertise to adequately review and assess the risks associated with this emerging research. As the stewards of health and safety at WFUSM, EH&S realized it had to quickly educate itself. Literature reviews were performed, and the importance of such education was brought before the various institutional committees. About this same time, a new Principal Investigator, who had research experience with nanomaterials and wanted to use nanomaterials, contacted EH&S.

In conjunction with this faculty expertise, a presentation was developed to explain nanoscience, and the environmental, health and safety concerns to the institutional committees. After taking in the presentation, the Chemical Safety Committee decided to form the Nanomaterials Subcommittee (the Subcommittee), which was tasked to develop health and safety guidelines for using nanomaterials in research. The Subcommittee began meeting monthly, creating momentum for the development of the draft guidelines.

Multiple resources were used to develop and refine the draft guidelines. The guidelines utilized NIOSH's *Approaches to Safe Nanotechnology* publication. Additional information was culled from the European Commission's NanoSafe project, the International Council on Nanotechnology's (ICON*) GoodNanoGuide*, AIHA, programs from peer institutions, and multiple research publications from various journals. In addition, EH&S staff attended the technical sessions on nanomaterials at the 2009 American Industrial Hygiene Association (AIHA) Conference and Exposition and the 2009 American Society of Safety Engineers (ASSE) Professional Development Conference and Exposition (PDC). The additional information had opposing effects on subsequent versions of the guidelines. For example, confirming that HEPA filters capture nanomaterials allowed EH&S to relax the ventilation requirements specified in the guidelines. However, new absorption information confirmed that EH&S needed to increase dermal protection and other PPE requirements in laboratories and animal research spaces.

The foundation for the guidelines is based on a hierarchical approach to control measures in WFUSM research and animal spaces. The Subcommittee convened to address the greatest concern: respiratory absorption by WFUSM personnel. The guidelines spell out that all free particulate nanomaterials should be manipulated in exhausted enclosures (i.e., fume hoods, glove boxes, and Class II Type A2, B1 or B2 biosafety cabinets). One challenge of specifying exhausted enclosures on the WFUSM campus is that ventilation and fume hood design can vary, depending on the research building. Variable Air Volume (VAV) fume hoods and hard-ducted Class II Type B2 biosafety cabinets were identified as engineering controls of choice that, unfortunately, limit the locations on campus where the manipulation of free nanoparticles can occur. The lack of VAV hoods and hard-ducted biosafety cabinets may become a challenge for the institution as nanomaterials research on campus swells.

The guidelines detail specific administrative controls and PPE requirements. Administratively, biosafety cabinets that are used for nanomaterial research will be certified semi-annually, instead of annually. Prefilters and HEPA filters used in biosafety cabinets will be changed and serviced by vendors using a bag in/bag out process to minimize personal exposure to any toxic substance the filters may have trapped. With regard to PPE, EH&S identified research showing that dry nanomaterials have the ability to pass through the woven fabric typical of lab coats worn throughout WFUSM labs. The Subcommittee determined that, if researchers will be using dry nanomaterials, they need to wear disposable Tyvek lab coats or disposable Tyvek sleeves and double nitrile gloves. Again, the added requirements of the guidelines create challenges for departments with limited budgets.

The Ugly

By ugly, unknown is meant. Even with a set of guidelines in place, numerous questions remained about how to categorize the risks associated with different nanomaterial research at WFUSM. How to communicate the hazard information regarding nanomaterial research to lab personnel, animal research personnel, and visitors? How to protect research and animal resources staff from nanomaterials that may be shed from research animals? Where to house larger animals that may be injected with and shedding nanomaterials? How to perform employee exposure monitoring to nanomaterials? Even if employees were monitored, are the PELs listed in Table 1 applicable for determining exposure to nanomaterials?

While the WFUSM General Guidelines for Handling and Working Safely with Nanomaterials provided the basic hierarchal structure for controlling nanomaterial hazards, the Subcommittee still lacked a method for categorizing the risks associated with the nanomaterial protocols submitted for review. Instead of creating an all new risk assessment and communication system, the Subcommittee decided to graft a system onto existing, proven frameworks. The system is called the Nanosafety Levels (NSL), and is a hybrid of the four biosafety levels described by the CDC and NIH in the fifth edition of the *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) publication and the Department of Homeland Security's Advisory System (colored threat levels).

Both the biosafety levels and the colored threat levels were familiar to researchers on the committee and employees working with nanomaterials. Essentially, the NSL system is a way to group nanomaterials into categories of risk based on the properties of the nanomaterial, the known health hazards associated with nanomaterials, and the hierarchy of controls used to minimize personal and environmental exposure. The Subcommittee decided to prioritize the characterization of specific types of nanomaterials, beginning with nanomaterials currently used on campus. Researchers at WFUSM procure nanomaterials from a range of sources, including lab supply companies, the Wake Forest Nanotechnology Center, or by creating the material themselves in their own labs. Therefore, the Subcommittee requested that researchers report the properties of the nanomaterials they use (or plan to use) to the Subcommittee to jumpstart the creation of the NSL levels. Ultimately, four levels were created, each with their own color, NSL 1-green, NSL 2 -yellow, NSL 3-orange, and NSL 4-red. The criteria used to categorize nanomaterials into the four NSL levels included form of nanomaterial and known knowledge of material. NSL 1-green nanomaterials present minimal hazards to employees and the environment and, therefore, require the least controls. NSL 4-red nanomaterials present substantial hazards to employees and the environment and, therefore, require stringent engineering, administrative, and PPE controls. The NSL chart can be seen in Figure 1.

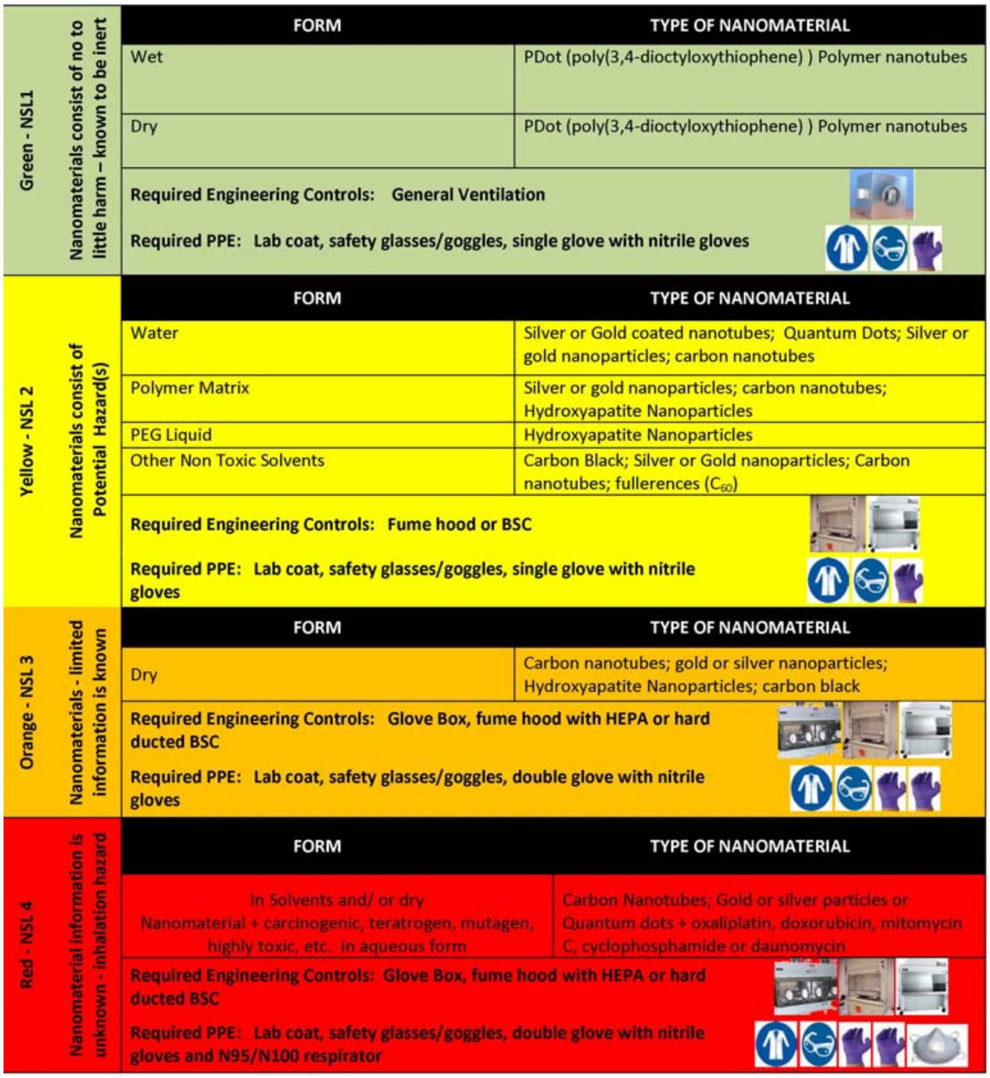

Figure 1. Nanosafety Level System

WFUSM laboratories have specific NSL levels, and Animal Resource Areas have separate, specific NSL levels. The Subcommittee leaned heavily on subject matter experts to make reasonable and prudent decisions when knowledge gaps existed for the health hazards of specific nanomaterials. The benefit of the NSL system is that any employee on campus can quickly identify the hazard based on the color codes. Whether you are a researcher working with nanomaterials or a member of the housekeeping staff, the color codes communicate the hazards associated with the nanomaterials used in lab or animal areas quickly and effectively. Since new information on the environmental, health, and safety of nanomaterials is generated quickly and evolves constantly, the Subcommittee's task continues. The guidelines and the NSL system are routinely reviewed, evaluated, and updated as new information becomes available.

Another unknown for the Subcommittee was the question of how to protect research staff and animal resources staff from nanomaterials that may be shed from research animals. The Animal Resources Program at WFUSM has several engineering controls available that would help to minimize personal exposure to nanomaterials. The engineering controls are animal-dependent and cater more towards small animals. Micro-VENT mouse racks are HEPA-filtered and create a closed ventilation system, effectively eliminating exposure to shed nanomaterials until the cages must be cleaned. In order to avoid exposure during cage cleaning, disposable cages could be utilized to house small animals. Unfortunately, switching to all disposable cages would be cost prohibitive for the institution, as well as wasteful. Reusable cages can be changed underneath a hard-ducted Class 2 Type B2 biosafety cabinet, but WFUSM only has a few of these hoods on campus. Complications will mount when nanomaterial research moves into larger animals that cannot be housed in ventilated cage rack systems and require far more hands-on husbandry. In addition to the engineering controls in place, several administrative controls were created to help reduce exposure to shedding animals and their bedding. Following a similar pattern established by the creation of the NSL, the Subcommittee once again grafted a system onto an existing, proven framework. EH&S creates cage cards for animal cages to communicate to animal resources personnel that the animal within the cage has been injected with a highly toxic material. The cage cards are used when animals have been treated with a carcinogen, mutagen, reproductive toxin, chemotherapeutic agent, and now, nanomaterials. The cage card system is integrated with the NSL of the nanomaterial, so the NSL level dictates the cage card needed on the housing of the animal (Figure 2).

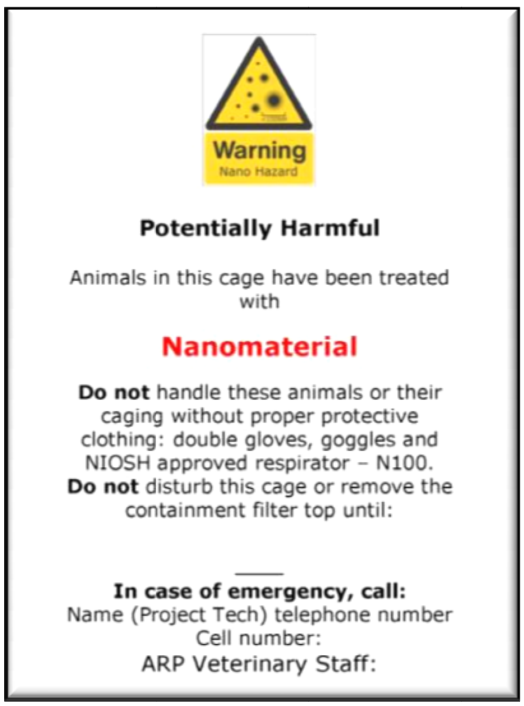

Figure 2. Cage Cards

Animal resource personnel and researchers know at a glance that the animal has been treated with nanomaterials and to be vigilant when handling, changing, and cleaning the cage and the bedding.

While the NSL and cage cards communicate the risk associated with specific nanomaterial research on campus, questions remained for the Subcommittee and EH&S on how to properly quantify personal exposure to nanomaterials in WFUSM laboratories and animal care facilities. NIOSH describes a proposed sampling strategy in its *Approaches to Safe*

Nanotechnology publication, but the strategy relies primarily on area sampling, which creates uncertainty when estimating worker exposure. If personal exposure could be quantified reliably, then which permissible exposure limit should be referenced? Lacking significant toxicological data and proven PELs, the Subcommittee and EH&S agreed to strive to keep exposures as low as reasonably achievable (ALARA), a longstanding exposure principle in radiation protection. Moving forward, EH&S plans to perform wipe sampling in labs, using nanomaterials to see if surface contamination remains after laboratory personnel have completed their procedures and cleaned the work surface. Samples would be analyzed by a Tunneling Electron Microscope (TEM). The wipe sampling will be a crude, qualitative method, but it should shed light on the relative hygiene of nanomaterial research and the cleaning methods of the labs. Wipe sampling may help to identify hot spots of contamination and improve administrative controls enacted with the approval of the researcher's chemical safety protocol. Beyond wipe sampling, EH&S will evaluate 45mm open-faced cassettes to perform personal sampling using NIOSH methods 7300 (metals) and 5040 (elemental carbon). EH&S will continue to refine area and personal sampling procedures as new techniques become available and best practices are established.

Conclusion

The nanotechnology field is quickly expanding and will create a large global industry in the coming years. As an academic research institution, WFUSM procures, creates, and researches nanomaterials on a laboratory scale. While the quantities may be smaller than those found in industry, the variety of nanomaterials on campus may exceed those found in industrial settings. In an effort to create prudent practices in the absence of federal or state regulations, WFUSM EH&S utilized an existing and familiar risk management system (chemical research protocols) and augmented and expanded the system for engineered nanomaterials. The expanded risk management approach included the development of general guidelines for working with nanomaterials safely on campus, the creation of a nanosafety level system, and the addition of nanomaterial cage cards. The nanosafety level system categorizes nanomaterials into risk groups that are color-coded and numbered to communicate the hazards of nanomaterials quickly and effectively. The nanomaterial cage cards are integrated with the NSL risk levels, and help animal resources personnel to quickly ascertain the risk associated with handling, cleaning, and disposing of animal caging and bedding. The Engineered Nanomaterials Subcommittee, formed to create the prudent practices introduced here, will continue to evaluate and update the guidelines and nanosafety levels as new health and safety information becomes available.

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Endnote

[1] Markiewicz, D. "Get the most out of Your Nanotech Learning Experiences." *The Monitor. 2009* Vol. 8, No. 3: 13 – 15 (retrieved January 15, 2010) (http://www.google.com/url?q=http://www.asse.org/nanotechnology/docs/Dan%2520Markiewicz%2520Article.pdf&ei=pa5QS4bFEceWtgfI3JmtDA&sa=X&oi=nshc&resnum=1&ct=result&cd=2&ved=0CAoQzgQoAQ&usg=AFQjCNHPkRgNT0TGbvmrvPueBL09UAOT_g).

Bibliography

American Conference of Governmental Industrial Hygienist. *2009 Guide to Occupational Exposure Values*. Ohio: ACGIH, 2009.

Balazy, A., Podgorski, A., and Gradon, L. "Filtration of Nanosized Aerosol Particiles in Fibrous Filters. I – Experimental Results." Abstracts of the European Aerosol Conference 2004. 2004.

Balazy, A., Toivola, M., Reponen, T., Podgorski, A., Zimmer, A., and Grinshpun, S. "Manikan-Based Performance Evaluation of N95 Filtering- Facepiece Respirators Challenged with Nanoparticles." *Annals of Occupational Hygiene.* 2006. Vol. 50, No. 3:259-269.

Best Practices for Handling Nanomaterials in Laboratories. Massachusetts Institute of Technology (MIT) (retrieved January 8, 2010) (http://web.mit.edu/environment/pdf/University_Best_Practices.pdf).

European Agency for Safety and Health at Work. 2009. EN8, *European Risk Observatory Report, Expert Forecast on Emerging Chemical Risks Related to Occupational Safety and Health* (retrieved January 8, 2010) (http://osha.europa.eu/en/publications/reports/TE3008390ENC_chemical_risks).

"Good Nano Guide," ICON hosted website on best EHS practices for nanomaterials (retrieved January 8, 2010) (http://goodnanoguide.org/tiki-index.php?page=HomePage).

"Interim Guideline for Working Safely with Nanotechnology." Texas A&M Engineering (retrieved January 8, 2010) (http://tees.tamu.edu/EPjsp/f_dwn.jsp?image_num=E6305E7017B57C42CC3529CA61C96C97).

Health and Safety Executive (HSE). March 2009. WEB38, "Risk Management of Carbon Nanotubes." (retrieved January 8, 2010) (http://www.hse.gov.uk/PUBNS/web38.pdf).

Knoweles, E. "Nanotechnology – Evolving Occupational Safety, Health and Environmental Issues." *Professional Safety,* Mar. 2006: 20-27.

Lee, KW. Liu, BYH. "Experimental Study of Aerosol Filtration by Fibrous Filters." *Aerosol Science and Technology*. 1981. 1:1:35-46 (retrieved March 27, 2009) (http://www.informaworld.com/smpp/content~db=all~content=a777854077).

McShane, B. "Nanotechnology – Is there Cause for Concern?" *Professional Safety,* Mar. 2006: 28-34.

Paik, S, Zalk, D and Swuste, P. "Application of a Pilot Control Banding Tool for Risk Level Assessment and Control of Nanoparticle Exposures." *Annuals of Occupational Hygiene* Jul. 2008: 419-428. (retrieved January 8, 2010) (http://annhyg.oxfordjournals.org/cgi/reprint/52/6/419).

"Potential Risks of Nanomaterials and How to Safely Handle Materials of Uncertain Toxicity." MIT Environmental Programs (retrieved January 8, 2010) (http://web.mit.edu/environment/ehs/topic/nanomaterial.html).

Rengasamy, S. King, WP. Elmer, BC. Shaffer, R. "Filtration Performance of NIOSH-Approved N95 and P100 Filtering Facepiece Respirators Against 4 to 30 Nanometer-sized Nanoparticles." *Journal of Occupational and Environmental Hygiene,* September 2008, 5:556-564.

Springston, J. "Nanotechnology – Understanding the Occupational Safety and Health Challenges." *Professional Safety,* Oct. 2008: 51-57.

Toxicology of Engineering and Incidental Nanoparticles. American Industrial Hygiene Association (AIHA). Teleweb Virtual Seminar. January 27, 2009.

U.S. Department of Health and Human Services, Center for Disease Control and Prevention, National Institute for Occupational Safety and Health (NIOSH). Publication 2009-125, *Approaches to Safe Nanotechnology Managing the Health and Safety Concerns Associated with Engineered Nanomaterials* (retrieved January 8, 2010) (http://www.cdc.gov/niosh/docs/2009-125/).

U.S. Department of Health and Human Services, Center for Disease Control and Prevention. *Biosafety in Microbiological and Biomedical Laboratories* (retrieved February 3, 2010) (http://cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc.htm).

Wang, H and Kasper, G. "Filtration Efficiency of Nanometer-Size Aerosol Particles." *Journal of Aerosol Science,* 1981, Vol 22, No. 1, pp.31-41.

Zalk, D, Paik, S and Swuste, P. "Evaluating the Control Banding Nanotool: A Qualitative Risk Assessment Method for Controlling Nanoparticle Exposures." *Journal of Nanoparticle Research,* Jun. 2009 (retrieved January 8, 2010) (http://www.springerlink.com/content/4t3241552m176137/).