Chemical Substances and Biological Agents

Studies and Research Projects

REPORT **R-656**

Engineered Nanoparticles

Current Knowledge about OHS Risks and Prevention Measures

Second Edition

Claude Ostiguy
Brigitte Roberge
Catherine Woods
Brigitte Soucy

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Legal Deposit
Bibliothèque et Archives nationales
2010
ISBN: 978-2-89631-478-2 (print format)
ISBN: 978-2-89631-479-9 (PDF)
ISSN: 0820-8395

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Note :

The following people have participated to the First Edition of this report published in 2006 by the IRSST in the Research and Studies Series, report # R-455. Some of the informations of this report have been kept, removed, modified, updated, or summarized in the current report.

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This publication is available free of charge on the Web site.

This study was financed by the IRSST. The conclusions and recommendations are those of the authors.

SUMMARY

Nanotechnology is a dynamic field and new products containing nanoparticles are being marketed every week. Already, more than 1000 are commercially available. This being the case, the development of these nanoparticles, their industrial preparation and their integration into different products involve a potential occupational exposure of hundreds of Québec workers. New Québec companies are introducing this field, thus increasing the number of potentially exposed workers. This trend should continue for several years.

An initial assessment of the state of scientific knowledge about the occupational health and safety aspects (OHS) related to synthetic nanoparticles (NP) was published by the IRSST in 2006 and covered the scientific literature until the end of 2004. What was found was that OHS knowledge was very fragmentary but that research in this field was rapidly growing. This current document aims to assess the state of current knowledge in this field and summarizes the data available until early 2010.

Overall, what emerges is that NP remain an important source of concern in OHS. In fact, not only does the diversity of commercially available chemical products of nanometric dimensions continue to increase, but also, the information available about the hazards specific to these substances is still very fragmentary. The literature gives us very little information specific to NP relating to their physical hazards like fires or explosions. As for health hazards, many toxicological studies on different substances have demonstrated toxic effects on various organs. It is found that in general, an NP will normally be more toxic than the same chemical substance of larger dimensions, but it is currently impossible to determine which measuring parameter for exposure is best correlated with the measured effects. The evaluation of occupational exposure must therefore address a series of different parameters, and the exposure data available are relatively rare. It should also be noted that at the present time, attention is particularly focused on carbon nanotubes (CNT), which seem to show, in different animal studies, toxicity similar to that of asbestos and consequently causing great concern in the international scientific community, mainly relating to prevention.

In a context of incomplete data for the majority of nanometric substances, it remains impossible to quantify the risks for workers in the majority of situations because the toxicity of the products, the level of dust contamination of workplaces, or their potential to cause fires or explosions remain not extensively documented or totally undocumented. Nevertheless, the majority of the means of exposure control for ultrafine particles should be effective against NP and much research is currently being carried out to confirm this.

In a context of uncertainty about the risks, and with an increasing number of potentially exposed workers, the current report paints a big picture of the OHS knowledge currently available in the NP field. In the absence of specific standards, a preventive and even a precautionary approach are recommended, and a review of the available means for minimizing worker exposure is presented. The needs for developing new knowledge are enormous and the authors propose some avenues of research that they feel are a priority in the current Québec context.

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1. CONTEXT AND OHS ISSUES

The field of nanotechnology (NT) is changing rapidly, and in the near future should have a profound effect on every sector of society. This is why a number of authors maintain that it constitutes a third industrial revolution, following the mechanization and computerization revolutions. As such, the industrialized countries -- quite rightly -- see the nano-world as showing great promise in economic development and spinoffs. Governments, as well as large firms, are developing strategic plans and investing considerable sums in research and development (R & D). Europe, for example, has made nanotechnology one of its seven priority areas in its targeted research, and has invested 1.3 billion Euros in it for the 2002-2006 period (www.cordis.lu/nmp/home.html)[1]. For the year 2008 alone, the *National Nanotechnology Initiative* (NNI) in the United States had a budget of 1.49 billion dollars (NNI, 2008). Canada, too, has invested in the design, manufacture and uses of NT (Chapter 5).

Yet what exactly are NT? The prefix *nano* signifies a billionth (10^{-9}). Thus, a nanometre (nm) is a billionth of a metre. Nanotechnology is concerned with the creation or handling of particles and materials that, as a minimum: (i) have one nanometric dimension, normally 1 nm to 100 nm, and (ii) are produced through the structured organization of groups of atoms and molecules, or through reduction, at the nanometric level, of macroscopic materials. The interest in NT is essentially based on the fact that certain particles reveal specific properties only when they are of nanometric dimensions. These properties may include chemical, biological, electronic, rheological, magnetic, optical (photon), mechanical and structural effects.

Although the development of NT is a modern multidisciplinary science, naturally produced and manmade materials of nanometric dimensions and exposure to particles of other dimensions of mineral or environmental origin, including the fine fraction of nanometric particles, have always existed. Some of the natural nanometric particles are of biological origin – including DNA with a diameter of around 2.5 nm and many viruses (10 to 60 nm) and bacteria (30 nm to 10 μm) — while others are found in desert sand, oil fumes, smog, and fumes originating from volcanic activity or forest fires and certain atmospheric dusts. Among those generated by human activity, we should mention diesel fumes, industrial blast furnace emissions and welding fumes, which contain particles of nanometric dimensions (Teague, 2004).

However, basing the definition of NP on their size may induce too broad an understanding of our field of research. It is therefore essential to set limits for the object of our study. In this report, the term ***nanoparticles*** (NP) will be used to describe particles from 1 to 100 nm, synthesized intentionally with an objective of commercial use of one of the unique properties at these dimensions.

Note that an ISO standard, which was at the ballot stage at the time of the final writing of this document, proposes a different terminology. It is suggested that the term *nanomaterials* be used, which includes nano-objects as well nanostructured materials, with the latter being non-nanometric objects but having a fine nanometric-dimension structure. Nano-objects consist of a) nanoparticles whose three dimensions are nanometric, b) nanofibres (nanowires, nanotubes and nanorods) having two nanometric dimensions, and c) nanoplates with only one nanometric

[1] At the time the report was in preparation, all the websites cited were in operation.

dimension. Despite this situation, the editorial committee for this document decided to retain the *nanoparticle* terminology in order to maintain continuity with the studies published earlier by the IRSST and so as not to confuse the readers of this document with new terminology, not yet in common use in the scientific literature. Therefore, the term *nanoparticle* (NP) used in this document refers only to synthetic NP and consists of all of the elements in the nano-object group proposed in this new ISO standard.

In the past few years, we have noted an awareness of the urgency of developing knowledge relating to synthetic NP risks for occupational health and safety (OHS) and for the environment and public health in general. To all indications, we currently have limited knowledge of OHS specific to NP, primarily due to a lack of information on toxic risks occupational exposure and working conditions. Even if it is also possible to measure exposure with the technologies currently available, our ability to predict the impact on workers' health is therefore very limited.

However, various NP are already coming onto the market after manufacturing. There are already more than 1000 commercialized products containing nano components. The manipulation of these materials in the work environments generates some concerns and raises questions regarding the risks to workers' health and safety. The purpose of this document is to report on our current knowledge.

The present document is intended primarily to serve as a source of information for anyone seeking to understand NP, what they are used for and, above all, the occupational health and safety issues related to their synthesis and handling. Thus, the information summarized here could be of interest to a wide variety of groups, including not only occupational health and safety professionals, but also researchers, evaluation- and risk-management specialists, employers and decision makers in industry, governmental agencies that develop policies, laws and regulations, as well as workers who may be potentially exposed and the general population wishing to find out more on the subject.

In order to limit the length of this document, some information already presented in the first edition will be summarized here, and the reader interested in more detailed information can consult the IRSST report (http://www.irsst.qc.ca/files/documents/PubIRSST/R-455.pdf), (Ostiguy *et al.*, 2006b).

2. OBJECTIVES

The principal objective of the study is to draw an overall portrait of knowledge on NP, emphasizing OHS and prevention aspects. In no way should this document be considered a critical analysis of the existing scientific literature. Amongst other things, we will describe (i) the principal known health risks and impacts, (ii) safety risks, (iii) the characteristics, exposure assessment and control of these risks, as well as OHS research needs relevant to the field of NT. The report also seeks to strengthen Québec expertise in the field. This expertise should provide effective support to research teams and institutions synthesizing or using these products.

3. METHODOLOGY

The initial literature review of available information up to the end of 2004 had allowed us to observe the rapid changes in knowledge occurring in the field of OHS dealing with NP. In light of the fact that the methodology employed at that time enabled us to reach our objective, it has been retained to produce the second edition, which covers the scientific literature up to early 2010.

Consequently, the present report is based on:

- analysis of the scientific literature that uses approaches commonly used for this type of research; these approaches are found in journals with peer committees. The IRSST Information Library and the CSST Information Resource Centre conducted the literature search. The principal data banks and search engines consulted were MedLine, Toxline, PubMed, Inspec, Coppernic, Embase, Ntis, Ei, Compendex, SciSearch, Pascal, Alerts, Teoma and Scirus. A number of keywords (in both French and English) were used, including nanotoxicology, nanotechnology, nanoparticle, nanomaterial, health effects, explosion, fire, toxicity and toxic. This information was particularly useful in documenting the toxicity of nanoparticles;
- an Internet search, primarily to document (i) the types of processes facilitating the manufacture of these products, and (ii) the description of these products, their properties and their potential uses;
- the use of scientific reviews by recognized committees of international experts in the fields of nanoparticles and nanotechnology;
- information collected from Quebeckers directly involved in the nanotechnology field;
- discussions with the editorial committee of the report.

4. TERMINOLOGY, CLASSIFICATION, PROPERTIES AND CHARACTERISTICS OF NANOPARTICLES

4.1 Terminology

As discussed previously, NP are products that have been synthesized because they have unique properties relating specifically to their dimensions. Moreover, in recent years substantial progress has been made in defining them, especially through the creation of groups of experts working in the area of standards development. Today, an international consensus exists. Recent studies by British Standards Institute (BSI, 2005), the American Society for Testing and Materials (ASTM, 2006), the Nordic group (Schneider *et al.*, 2007) and the ISO (2008a) have defined NP as particles with one or several dimensions of 1 to 100 nm. By contrast, terminology surrounding various aspects of NT continues to be the subject of debate as described in chapter 1.

One-dimensional systems, such as thin films or manufactured surfaces, have been used for decades in the electronics, chemical and engineering industries. Production of thin or monolayer films is now common practice in the electronics field, as is the use of made-to-measure surfaces for solar cells or catalysis. These fields are known and the risks are normally well controlled. The properties of two-dimensional systems (carbon nanotubes (CNT), inorganic nanotubes, nanowires, nanofibrils and biopolymers) are less well understood and the manufacturing capabilities are less advanced. Finally, some 3D systems are well known, such as metal oxides, carbon black, titanium dioxide (TiO_2) or zinc oxide (ZnO), particles, precipitates, colloids and catalysts of nanometric dimensions, while others, such as fullerenes, dendrimers and quantum dots, currently pose major challenges in terms of production and understanding of their properties.

The question of NP dimensioning is crucial, because the properties of nanostructures are directly related to those of individual molecules instead of those of bulk materials (Kohler and Fritzche, 2004; Royal Society and Royal Academy of Engineering, 2004). In fact, NP are observed to have radically different, even unique properties at nanometric dimensions: exceptional strength, programmable electrical conductivity, unsuspected optical properties, etc. The very principles of chemistry and classical physics of solid materials must be replaced by quantum approaches based on the probabilities that each atom, each molecule, can play a determining role and that the interactions among them have a decisive impact on the behaviour of the whole. Thus, the classical mechanical parameters of solids no longer prevail. In fact, it is the individual molecular and atomic dimensions and interactions that determine the arrangement, stability, flexibility and function of nanostructures.

Two central factors seem to be responsible for the changes in properties observed in NP: a much larger relative surface per unit of mass and a predominance of quantum effects. The first factor is responsible for the changes in reactivity, which may increase considerably with a decrease in the size of NP. The second factor, observed for certain particles of a few nm or tens of nm, induces changes in optical (photonic), electrical, electronic, mechanical, chemical, biological, rheological, structural or magnetic properties.

Once the dimensions are determined, a simple way to classify NP is to group them by chemical composition, and this is the approach we will use. However, we should mention that many NP are coated immediately after synthesis to prevent any aggregation and preserve their properties. These modifications, in return, may have a direct impact on their composition, size and reactivity.

4.2 Classification and Properties

In view of the objective of our report and the fact that production of new nanomaterials is a fast-growing field of research, only the most common products will be described briefly. For more details, it is recommended that you consult the special edition of the Journal of Materials Chemistry regarding preparation of nanomaterials (Rao, 2004). Rao *et al*. (2004) have also published a volume describing the theory, synthesis, properties and applications of these products. Many specialized journals have also emerged in the last decade and new NP structures with novel properties are reported regularly in these publications. The reader can also consult the first edition of the IRSST report (http://www.irsst.qc.ca/files/documents/PubIRSST/R-455.pdf; Ostiguy *et al.*, 2006b).

4.2.1 The Main Carbon-Based Nanoparticles

4.2.1.1 Fullerenes

Fullerenes are spherical cages composed of carbon atoms, which are bound to three other atoms in sp^2 hybridization (Figure 1). The most widely studied form, synthesized for the first time in 1985 (Kroto *et al*.), is spherical and contains 60 carbon atoms, C_{60}, although there have been reports of structures containing 28 to 1500 carbon atoms, which can reach a diameter of 8.2 nm. Formation of multilayer fullerenes has also been reported, with dimensions potentially ranging from 4 to 36 nm (Sano *et al.*, 2002).

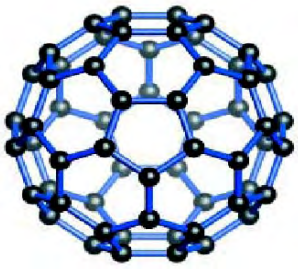


Figure 1 : Schematic Representation of a Fullerene C_{60}

Given their interesting properties, it has been suggested that they be used in the electronic field, in batteries, solar cells and combustion cells, for data storage or gas storage, or as additives in plastics. Incorporating them into carbon nanotubes (CNT) modifies the electrical behaviour of fullerenes, creating regions with varying semiconductive properties, thus offering potential applications in nanoelectronics. Their optical properties vary according to wavelength, thus finding applications in telecommunications. Since fullerenes are empty structures with dimensions similar to several biologically active molecules, they can be filled with different substances and find medical applications, particularly in the therapeutic field against cancer or AIDS.

4.2.1.2 Graphene Nanofoils

Graphite is composed of a series of superimposed layers of a hexagonal network of carbon atoms, in which each atom is bound by three neighbouring carbon atoms in a planar network. Separating these layers into a monolayer around one nanometre thick makes it possible to obtain graphene foils. This allows unique electronic, magnetic, optical and mechanical properties to appear. Applications are currently envisioned in recovery and in the flexible electronic components field.

4.2.1.3 Carbon Nanotubes

Carbon nanotubes (CNT) are a new crystalline form of carbon. Wound in a hexagonal network of carbon atoms constituting a graphene nanofoil, these hollow cylinders can have diameters as small as 0.7 nm with lengths that can range from a few micrometres, and reach several millimeters in length (Hett, 2004a). Each end can be opened or closed by a fullerene half-molecule. These nanotubes can have a single layer (SWCNT for *single walled carbon nanotube*) or several layers (MWCNT for *multi walled carbon nanotube*) of coaxial cylinders of increasing diameters in a common axis. Multilayer carbon nanotubes can reach diameters of 100 nm (Aitken *et al.*, 2004). Endo (1986) seems to have been the first to report the existence of CNT.

They display metallic or semiconductive properties, depending on how the carbon leaf is wound on itself. The substitution of carbon atoms allows modification and adjustment of electronic properties. The current density that a nanotube can carry is extremely high and can reach one billion amperes per square metre (Pautrat, 2003, Aitken *et al.*, 2004). Light and flexible, chemically very stable and totally insoluble, mechanical resistance under tension, CNT is more than sixty times greater than that of the best steels, even though they weigh six times less (Aitken *et al.*, 2004). They also present a very large specific surface area, and a great capacity for molecular absorption (Maynard, 2004). The different processes of synthesis, purification and post-synthesis treatment will lead to products displaying very different properties (Helland *et al.*, 2007). CNT are ranked among the most promising materials and researchers have detected a whole range of potential CNT applications in nanotechnology: polymer composites, electron transmitters, electromagnetic armouring, supercapacitors, storage of gases, including hydrogen, batteries, structural composites (Eklund *et al.*, 2007).

4.2.1.4 Carbon Nanofibres

Just like CNT, carbon nanofibres are composed of graphene foils. But unlike nanotubes, they do not wind into a regular cylinder. Instead they form a cone-shaped or cup-shaped structure. In

view of these particularities, the mechanical and electrical properties of nanofibres will be different than those of CNT. Their use is envisioned as additives in polymers, as catalytic media and for gas storage.

4.2.1.5 Carbon Black

Carbon black is composed of partially amorphous graphitic material, with a substantial fraction of the elementary particles of nanometric dimensions, generally from 20 to 70 nm. These particles, mostly spherical, are bound in aggregates that interact strongly with each other to form agglomerates of up to 500 nm. These are often marketed in the form of pastilles or blocks and have been used in very high volumes for the past few decades, mainly as pigments and strengthening agents in rubber, particularly for tires. This technology is well known, and it is only partly covered in the report.

4.2.1.6 Carbon Nanofoams

Carbon nanofoams are the fifth known allotrope of carbon, after graphite, diamond, carbon nanofibers and fullerenes. In carbon nanofoam, islands of carbon atoms, typically from 6 to 9 nm, are randomly interconnected to form a very light, solid and spongy three-dimensional structure, which can act as a semiconductor. Carbon nanofoams display temporary magnetic properties (Health and Safety Executive, 2004b).

4.2.2 Other Inorganic Nanoparticles

4.2.2.1 Metals

Most metals have been or can be produced in nanometric dimensions. Among them, gold NP are studied in particular and show an optical resonance spectrum in the visible range, which is sensitive to the environmental conditions, size and shape of NP. Their unique properties make it possible to envision a series of applications, particularly as optical markers for medical diagnosis or as cancer treatment agents. Nanometric silver is also produced in large quantities and is mainly used for its antimicrobial properties (ICON 2008; Hansen 2009). Nanometric platinum, palladium and rhodium are used in catalytic converters, iron, nickel and cobalt as catalysts, particularly for the synthesis of carbon nanomaterials, aluminium as a fuel, iron as a doping metal and copper in electronics. Gold, copper, silicon and cobalt nanowires, capable of being electrical conductors or semiconductors, have also been perfected and could be used to transport electrons in nanoelectronics. Finally, nanowires have been developed based on different metals, oxides, sulphides and nitrides.

4.2.2.2 Metal Oxides

Several metal oxides of nanometric dimensions have been created, but the most common ones, because they are produced on a large scale, are probably silica, titanium dioxide and zinc oxide. They are used either natural or coated, mainly in the fields of rheology, plastics and rubbers as active agents and additives (SiO_2), in sun creams (TiO_2, ZnO), and as pigment in paint (TiO_2). Different metal oxides have appeared in varied forms: nanotubes, nanorods, nanoflakes, etc. In addition, certain structures show interesting properties for virtual applications in fields such as

sensors, optoelectroics, transducers and medicine… Other metal oxides are also produced, including cerium, iron, copper, zirconium, aluminium, nickel, antimony, yttrium, barium and manganese oxides, as well as nanoclays (ICON 2008).

4.2.2.3 Quantum Dots

An important field of research, for the past fifteen years, quantum dots are typically composed of combinations of Group II and IV elements or Group III and V elements of the periodic table. They have been developed in the form of semiconductors, insulators, metals, magnetic materials or metallic oxides. The number of atoms in quantum dots, which can range from 1,000 to 100,000, makes them neither an extended solid structure nor a molecular entity (Aitken *et al.*, 2004). With smaller dimensions than the exciton Bohr radius, they display unique optical and electronic properties at diameters of about one to ten nm. Because of their quantum confinement, they can, for example, absorb white or ultraviolet light and reemit it at a specific wavelength a few nanoseconds later (Aitken *et al.*, 2004). Depending on the composition and size of the quantum dot, the light emitted may range from blue to the near infrared.

The flexibility of quantum dots and their associated optical properties make it possible to envision applications in fields such as multicolour coding in the study of genetic expression, in high-resolution and high-speed screens and in medical imaging. Their high surface-to-volume ratio allows them to be combined with antibodies, proteins and oligonucleides (Michalet *et al.*, 2005). Some quantum dots are modified to produce drug vectors, diagnostic tools and inorganic solar batteries (Akerman *et al.*, 2002; Michalet *et al.*, 2005).

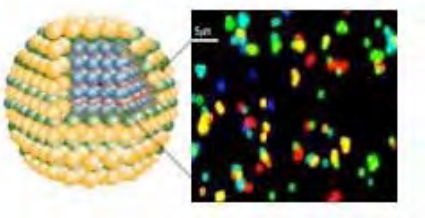


Figure 2 : Schematic and Visual Representation of a Quantum Dot

4.2.3 Organic Nanoparticles

4.2.3.1 Organic Polymers

Many common organic polymers can be produced in nanometric dimensions. The polyvinyl chloride or latex thus produced, for example, can be solubilized or modified chemically under certain conditions. Some of these organic polymers can be prepared in the form of nanowires, resulting in their use in the development of liquid-phase or gas-phase ultrafiltration systems, or particularly as sensors. Some biodegradable organic nanofibres could be used in medicine for tissue reengineering or bone regeneration or to control the release of drugs.

New structures have also been synthesized, such as dendrimers, which represent a new class of controlled-structure polymers of nanometric dimensions. These dendrimers are synthetic three-dimensional macromolecules developed from a monomer, deploying and multiplying new

branches in successive tiers, until a synthesized symmetrical structure is constituted (ICON, 2008). They are considered to be basic elements for the large-scale synthesis of organic and inorganic nanostructures of dimensions ranging from 1 to 100 nm and exhibit unique properties. Dendrimers allow precise, atom by atom control of nanostructure synthesis, based on the dimensions, shape and desired surface chemistry. Given that they can be designed as hydrophilic or hydrophobic, their use can be highly diversified. With different reactive surface groupings, it is envisioned that dendrimers will be used extensively in the medical and biomedical field as a means of delivering drugs and nutraceutics, as therapies, for biotests or as imaging contrast agents (Tomalia, 2004; ICON, 2008). Compatible with organic structures such as DNA, they can also be manufactured to interact with metal nanocrystals and nanotubes or be encapsulable or show unimolecular functionality (Tomalia, 2004). Their use is also anticipated as an ink in printers, as a metallic chelating agent in ion exchanger resins, in coatings, in cosmetics or as a viscosity modifier or environmental rehabilitation substance (ICON, 2008).

4.2.3.2 Biologically-Inspired Nanoparticles

Biologically inspired NP are highly diversified but normally include structures in which a biological substance is encapsulated, trapped or absorbed on the surface. In particular, lipids, peptides and polysaccharides are observed, used as vector for the targeted transport of drugs, receptors, medical imaging chemicals or nucleic acids. For example, liposomes, micelles or polyplexes are found, some of which may come from natural materials, while others are synthesized. These structures, widely studied in the medical and pharmacological field, will not be discussed in this document, given the lack of information available regarding them in the scientific literature dealing with OHS.

5. DEVELOPMENT, PRODUCTION AND USE OF NANOPARTICLES

The first edition of this report (Ostiguy *et al.*, 2006b) covered the important aspects until the end of 2004. In this publication, only the most recent or most relevant information is presented. NT is currently one of the most dynamic research fields in the world and is emerging as a strategic priority in several industrialized countries. A more recent assessment, by a group of experts from Nordic countries (Schneider *et al.*, 2007), forecast that it would reach US$2.6 billion in 2015, while Lux Research (2007) concurred with this figure, but said it would reach it in the year 2014 instead. In addition, in a 2006 report, the French Agency for Environmental and Occupational Health and Safety (AFSSET) expects that the principal economic impact of NT will occur in the nanomaterials sector, electronics and the pharmaceutical industry (Figure 3, derived from AFSSET, 2006). The creation of the US Nanobusiness Alliance, the Europe Nanobusiness Association and the Asia-Pacific Nanotechnology Forum, whose shared objective is to commercialize nanoproducts, is a good illustration of both the significance attributed to these markets and of international competition in the field. Furthermore, Québec is moving in the same direction by way of NanoQuébec. In its 2006-2007 Annual Report, NanoQuébec reported annual sales of nanoproducts by Quebec firms of less than $2 million for the year 2005, of about $8 million for 2006, while forecasting sales of over $25 million for 2008. More recent data are not available. The NanoWerk database (www.nanowerk.com) has already claimed (in March 2009) that there are 2225 NP available from 142 suppliers, while the site http://nanotechproject.org/18/esh-inventory provides a list of R & D projects in the OHS of NT.

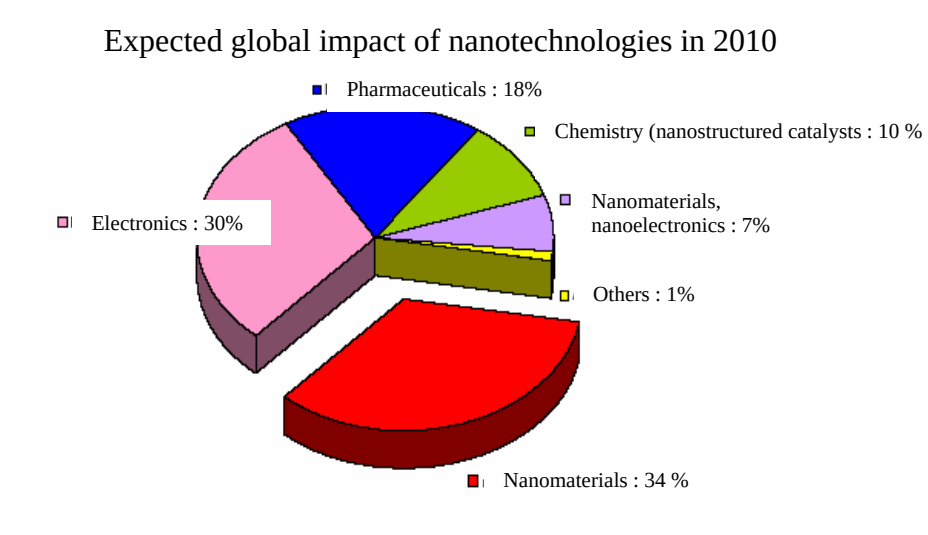


Figure 3 : Percentage Distribution of the Expected Global Economic Impact of Nanotechnologies in 2010

5.1 Efforts in Research and Development[2]

Research efforts at the global level

Research efforts at the global level were evaluated at over US$9.6 billion for the year 2005 alone (Lux Research Inc, 2007; Helland *et al.*, 2007). Despite these colossal investments in the development of commercial applications, research in the OHS sector, though stepping up the pace, is still limited. For example, in a report published in 2008, Rejeski states that in 2006 the budget spent by the National Nanotechnology Initiative (NNI), and incorporating Environmental Protection Agency (EPA), National Institute for Occupational Safety and Health (NIOSH) and National Institute on Health (NIH) expenditures dedicated completely to NT risk assessment, were of the order of $13 million, or about 1% of the NNI annual budget. For 2009, the budget requested by the NNI is US$1.527 billion (Roco, 2009), of which $76.4 million is devoted to environmental issues and OHS.

Research efforts in Asia

While several Asian countries have invested in nanotechnology research, Japan is the principal actor in the field and is making a major research effort in the field. Obadia (2008) ranks Japan second in the world, following the United States, with South Korea in fourth place. In addition, Japan has adopted an integrated development policy in this field. In addition, it already produces a significant volume of nanotechnology and the Japan National Institute for Occupational Safety and Health (JNIOSH) is already intensely involved in OHS research.

Research efforts in Europe

For the period 2002-2006, the Sixth EU Framework Programme for Research and Technological Development (FP6) of the European Research Area (European Commission, 2002), has funded the carrying out of 550 projects valued at 1.4 billion Euros, 28 million of which are dedicated to OHS research (European Commission, 2007). The program is designed to develop a European research and communication network incorporating all aspects of NT, including business and scientific aspects, and information intended for the general public. Following extensive consultation with its members, the Seventh EU Framework Programme for Research and Technological Development (FP7) (Commission of the European Communities, June 2005), suggested increasing EU investments in R & D to strengthen the European position internationally. To these European efforts, we need to add the individual initiatives of several countries in the Union. The most committed of these countries are Germany, Great Britain, France, Switzerland, Belgium and the Netherlands. Germany is ranked third in the world in research (Obadia, 2008).

Europe also invests in research on the risks associated with nanomaterials and NT, and does so at both the European Commission level and the individual government level. We should mention in particular the NanoImpactNet network, which consists of 24 European research centres as well as some centres outside Europe with the objective of harmonizing research on NT and of sharing OHS knowledge (www.nanoimpactnet.eu).

[2] More detailed information is available in Ostiguy *et al.*, 2006b

For its part, the British government mandated the Royal Society, the National Academy of Science and the Royal Academy of Engineering to supervise a study on the benefits and risks of NT, and has published several of its reports. In an Institute of Occupational Medicine (IOM) report, Aitken *et al.* (2009) drew up a balance sheet of the research and published a substantial list of research recommendations, just like the Council for Science and Technology (2007) had done with regard to the progress and commitments made by the British government and the contribution of Great Britain at the international level. In Sweden, a study of the same type was entrusted to Lund University, while in Germany several initiatives have been taken by the Parliamentary Office for Evaluation of Scientific and Technological Choices. In Switzerland, risk assessment has been carried out by the European office of technology assessment and the Swiss federal Institute of Technology. Furthermore, still in Switzerland, an inventory of nanoproducts used in the industrial environment was recently published (Schmidt and Riediker, 2008). Numerous research programs dealing specifically with OHS are underway in Europe.

Research efforts in North America

In the United States, the National Nanotechnology Initiative (NNI) seeks to accelerate the development of new knowledge in nanotechnology and facilitate its integration into commercially viable technologies (Mamberger and Kvamme, 2008; Roco, 2009). Created in 2000 by the federal government of the US, this agency supports a major multidisciplinary research program on NT, which has prompted Obadia (2008) to rank them as the world leader in research.

When it was created in 2000, the NNI's annual budget was US$422 million (Mamberger and Kvamme, 2008). Since then it has continued to grow, and for 2009 reached $1.527 billion (NSTC, 2008; Roco, 2009). In 2007, the Department of Defence received the highest proportion of subsidies, $450.2 million; it was followed by the National Science Foundation, which received $388.8 million, and the Department of Energy, with $236 million (Mamberger and Kvamme, 2008). Also, many American states invest in nanotechnology R & D. It is estimated that in the US the private funds invested in this area of R & D are equal to their publicly funded counterparts (Waters, 2003).

Many institutions are currently involved in research on NP toxicity and safe work methods. Among these, the National Institute for Occupational Safety and Health (NIOSH) assumes a leadership role in OSH, conducting and subsidizing NP research projects to improve our understanding of its health effects and develop safe methods for working with it. The National Science and Technology Council (2008), working for the NNI, recently presented its global strategy for integrating research related to the environmental, health and safety aspects of NT. Among other things, it expects that the National Institute for Standards and Technology (NIST) will be in charge of the section dealing with instrumentation, metrology and analytical methods. The National Institute for Health will develop knowledge on health risks, while the Environmental Protection Agency, the US EPA (2007a), will examine environmental aspects. Lastly, NIOSH will assess environmental exposure, while the Food and Drug Administration (FDA) and the EPA will develop risk management techniques. For 2009, the budget devoted to environmental aspects and OHS reach $76.4 million (Roco, 2009).

Research efforts in Canada

Through the National Research Council (NRC), Canada, too, has recognized the significance of NT, and in 2001 founded the National Institute for Nanotechnology (NINT) with premises of 10,000 m^2 and state-of-the-art facilities on the site of the University of Edmonton. The Government of Alberta invested substantial amounts in research and built large facilities for biopharmaceutical technologies (Feigenbaum *et al.*, 2004). Although several Canadian provinces have nanotechnology R&D groups and that a few developed coordination organizations (NanoBC, NanoAlberta, etc.), at the moment, none of the approaches taken at the provincial level seems as well structured as that employed by NanoQuébec.

Furthermore, the federal government developed an overall global strategy with a view to coordinating all research at the national level. To this end, it created the NINT Business Development Office (BDO) as a coordination centre designed to facilitate growth in the innovative capability of the Canadian industry as well as its garnering of market share in the new NT. Several federal departments had demonstrated great interest in NT. Among these institutes, Environment Canada, Health Canada, Natural Resources Canada and various NRC institutes developed R & D programs covering a broad section of NT (CNRC, 2009). Lastly, the Natural Sciences and Engineering Research Council (NSERC), the Canadian Institutes of Health Research (CIHR) and the Canada Foundation for Innovation (CFI) have funding programs for applied research in the field of nanotechnology.

Research efforts in Québec

For its part, Québec decided to develop a nanotechnology action plan, especially following the publication of a report by the Conseil de la science et de la technologie [science and technology board] entitled “Nanotechnology: mastering the infinitely small” and a general survey of the NT research carried out in 2001 by Lebeau. Quebec has also established NanoQuebec, which has a mandate to foster the development and enhancement of nanotechnologies in targeted priorities fields and make them an economic and social growth vector. All the major Quebec universities have active nanotechnology researchers (about 200 divided among about fifty teams); NanoQuebec posts a detailed directory (http://nanoquebec.ca/nanoquebec_w/site/index.jsp) of Quebec nanotechnology researchers and businesses on its web site. A more detailed description of Québec’s main actors is presented Chapter 12. In the field of OHS, the IRSST has already produced a variety of summary documents reporting on scientific knowledge in the field (Ostiguy *et al.*, 2006a, 2006b, 2008). In addition, it has now made available a Best Practices Guide, intended both for researchers and companies (Ostiguy *et al.*, 2009). The guide is designed to support the implementation of effective preventive measures with a view to fostering the safe development of NT in Québec.

Lastly, the IRSST, in collaboration with NanoQuébec, called for tenders to support the development of new expertise, by way of research, in the field of NP. The four projects it subsidized cover aspects related to metrology, evaluating occupational exposure, evaluating the effectiveness of control measures. Research results should be available by mid-2011.

5.2 Manufacturing Processes

Nanoparticles can be synthesized by different approaches. Nanoparticle production can be generally categorized into the bottom-up and top-down methods. In the bottom-up approach, nanoparticles are constructed atom-by-atom or molecule-by-molecule. In the top-down approach (*top-down*), a large structure is gradually underdimensioned, until nanometric dimensions are attained after application of severe mechanical stresses, violent shocks and strong deformations. The two approaches bottom-up and top-down tend to converge in terms of dimensions of the synthesized particles. The bottom-up approach seems richer, in that it allows production of a greater diversity of architectures and often better control of the nanometric state (relatively monodispersed granulometric sizes and distribution, positioning of the molecules, homogeneity of the products). The top-down approach, although capable of higher-volume production, generally makes control of the nanometric state a more delicate operation.

In this document, only the approaches most commonly used and likely to subject the works to high exposure during large-scale production of nanoparticles are discussed. The British Standards Institute (BSI, 2005) inventories no less than 29 major approaches for NP synthesis, while CRC Press recently published a handbook of NP preparation processes (Busnaina, 2007).

It is possible to categorize the synthesis processes according to the different mechanisms responsible for their formation (Aitken, 2004; BSI, 2005). Some authors prefer to divide the synthesis processes into three major categories based on the approaches used (synthesized NP) (AFSSET, 2006): chemical processes, physical processes and mechanical processes. ICON (2008) prefers to describe the different synthesis processes by major NP class. Processes are presented according to the AFSSET approach.

- Main chemical processes:
  - Vapour phase reactions (carbides, nitrides, oxides, metallic alloys, etc.);
  - Reactions and precipitations in liquid media (most metals and oxides);
  - Reactions in solid media (most metals and oxides);
  - Sol-gel techniques (most oxides);
  - Supercritical fluids with chemical reaction (most metals and oxides and some nitrides);
  - Chemical coprecipitation or hydrolysis reactions (metals and metallic oxides);
  - Chain and step polymerization in liquid phase (organic polymers, such as dendrimers and dendrons);
  - Gas phase polymerization, grafting, electrospinning (organic polymers, such as dendrimers and dendrons).

- Main physical processes:
  - Evaporation / condensation under inert or reactive partial pressure (Fe, Ni, Co, Cu, Al, Pd, Pt, oxides);
  - Laser pyrolysis (Si, SiC, SiCN, SiCO, Si_3N_4, TiC, TiO_2, fullerenes, carbonated soots, metallic oxides, etc.);

- Plasma synthesis or electric arc methods (metallic oxides);
- Combustion flames (metallic oxides);
- Supercritical fluid without chemical reaction (Materials for vectorization of active principles);
- Microwaves (Ni, Ag);
- Ionic or electronic irradiation (Production of nanopores in a material of macroscopic dimensions or nanostructures immobilized within a matrix);
- Low-temperature annealing (Complex metallic and intermetallic alloys with three to five basic elements (Al, Zr, Fe);
- Thermal plasma (Ceramic nanopowders, such as carbides (TiC, TaC, SiC), silicides ($MoSi_2$), doped oxides (TiO_2) or complexes (HA, YIG, perovskites);
- Physical deposit in vapour phase (TiN, CrN, (Ti, Al)N deposits, in particular).

o Main mechanical processes:

- Mechanosynthesis processes and mechanical activation of powder metallurgy processes (all types of materials (ceramics, metallic materials, metallic oxides, polymers, semiconductors));
- Consolidation and densification;
- Strong deformation by torsion, lamination or friction (metallic oxides).

The reader interested in more details on the synthesis processes is invited to refer to the following documents (BSI, 2005; Ostiguy *et al.*, 2006; AFSSET, 2006; Busnaina, 2007; ICON, 2008).

5.3 Applications of Nanotechnologies by Sectors of Activity

The number of products containing nanomaterials is increasing quickly and more than 1000 products are already commercially available. Some websites inventory these products:

- www.nanotechproject.org/44/consumer-nanotechnology
- www.nano.gov/html/facts/appsprod.html
- www.wilsoncenter.org/index.cfm?fuseaction=news.item&news_id=173868
- www.nanotechproject.org/inventories
- www.azonano.com/Applications.asp
- www.azonano.com/Industries.asp?Letter=_.
- http://www.nanowerk.com/nanobusiness/nanobusiness.php

NP will increase the performances of existing materials in many economic activity sectors (Roszek *et al.*, 2005; Ostiguy *et al.*, 2006b; AFSSET 2006; Faunce 2007; Kuzma, 2007). Table 1 presents, for different economic activity sectors, a few examples of performances sought through the use of NP.

Table 1: Examples of properties sought through the use of nanoparticles in different economic activity sectors

Automotive, aeronautical and spatial transportation	NP increase engine performance, mechanical and thermal resistance of materials, and energy performance, improve passenger safety and comfort, produce self-cleaning materials. NP reduce corrosion and pollution, lighten different aircraft components, costs, and fuel consumption. Development of ice and structure defects detectors.
Electronics and Communications	Development of high-density memories and miniaturized processors, very high brightness and definition flat screens, the availability of new polymers and new nanostructured composites allows the development of flexible electronic components and electronic paper.
Chemical and Materials Industry	Development of ceramics, pigments, powders, more effective multipurpose catalysts or production of lighter and stronger wires, corrosion inhibitors, multifunctional layers (thermal insulation, antiadhesive, antistatic), photoactive and self-cleaning paints, windows and clothings, and preparation of membranes for separation of materials (water treatment, dialysis), structured catalysts, ultraresistant coatings and extremely hard and resistant cutting tools.
Health and pharmaceuticals, biomedical, biotechnology and cosmetics industries	Development of new approaches for more effective, better targeted medical diagnostic (fluorescent markers, increased contrast through optical imaging, better characterization of certain parameters), and better targeted, more effective medical treatment, microlaboratory (lab-on-chips, cell-on-chips) and highly sensitive minisensors. In the cosmetic field, NP can improve optical properties (protection against ultraviolet radiation), resistance (water-resistant sun creams), brightness and transparency of products and the development of new anti-wrinkle, anti-aging and antibacterial product lines.
Agriculture	Development of solubilization modes on demand, better absorption for pesticides, fertilizers and other agricultural chemicals, optimization of livestock production through growth hormones and vaccines on demand, detection of pathogens in animals and plants.
Energy	Improvement of the performance of production systems and energy use, storage of hydrogen, creation of a new generation of photovoltaic cells, batteries and combustion cells, optimization of wind energy efficiency, smart windows, thermal barriers and more efficient insulating materials.
Manufacturing sector	Design various equipment to produce NP, incorporate them into value-added products and ensure characterization and production quality. Precision engineering for production of new generations of measuring instruments, development of new processes and new tools to manipulate matter on the atomic level, and development of apparatus capable of producing NP safely.
Environment and ecology	Sensors capable of real-time detection of multi-substances; reduction of polluting emissions, depollution of contaminated sites, treatment of effluents, protection of sensitive organisms and reduction of CO_2 emissions, production of ultrapure water, recovery and recycling of existing resources, improvement of decontamination and recycling of heavy metals, environmental monitoring.
Process and product safety	Real-time sensors and quality control on the atomic scale; protection against copying, anti-fraud security papers; marking processes for traçability.
Defence	Production of chemical and biological agent detectors, and the development of more powerful explosives, stealth systems; light, efficiently performing, self-repairing textiles, miniaturized surveillance systems and more precise guidance systems.
Rubber and plastics	Boost the performance of pneumatic tires, reduce their weight, increase their life cycle, favour their recycling, and reduce their noise emissions; eliminate the development of static electricity.
Metallurgy	Improvement of the properties of metals, reduction of friction and the use of lubricants in parts manufacturing; harder, more abrasion-resistant and corrosion-resistant cutting tools, and improvement in machining performance.

5.4 Nanotechnology Applications by Nanoparticle Type

Table 2 gives a few examples of properties and some specific NP applications.

Table 2 : Some Examples of Nanoparticle Applications

Nanoparticle type	New properties	Applications
C_{60} fullerenes	High electronic affinity	Improved magnetic properties, catalysts, pyrolysis, lubricants, solar cells, electrolytic membranes, ion exchange membranes, oxygen and methane storage, drug vectors.
TiO_2	Anti-UV optical properties and transparent to visible light, photocatalytic effect	Solar cells, anti-UV sun creams, anti-UV paint, environmental treatment, transparent wood surface treatment, self-cleaning material, antimicrobial agent.
Quantum dots	Colorimetric and electronic properties can be precision adjusted	Colourings, nanoelectronics and quantum computer, medical imaging, medical therapy, solar cells, catalysts.
NTC and inorganic nanotubes (e.g., molybdenum disulphide)	Good electric conductor Greater mechanical strength	Nanoelectronics and quantum computer, ultra-strong materials, static electricity dissipator, hydrogen storage, biosensors, chemical sensors, electromagnetic armouring, supercapacitors, reinforced polymer composites, super-strong cables, textiles, extremely light parts for land, air and space vehicles, additives.
Polymers/glasses	Miniaturization of chemical reactions	Lab-on-chip.
Liposomes	Biodegradable components	Drugs delivered to the action site, veterinary use.
Nanocapsules	Hollow shell	Medical applications, targeted drug delivery.
Photonic materials	Adjustable light transmission	Telecommunications, optical computers.
Nanomagnetic materials	Improved magnetic properties	Data storage.
Metallic oxides (Zn, Fe, Ce, Zr)	Large surface, optical properties	Ceramics, scratch-proof lens coatings, use in certain cosmetics and sun creams.
Nanoclays	Catalysis, strength, hardness, heat resistance and fire resistance	Oil refining, alters properties of composites and materials, flame retardant, mechanical reinforcement, rubber additive.
Carbon black	Large surface	Rubber and paint industries, inks.
Silica fumes	Rheological properties	Special concretes (durable, high strength, self-compacting, low density, low permeability) and higher quality used in construction of bridges, roads, marine structures, parking facilities, water purification and distribution systems; ceramics industry, mortars, plastic and rubber additive.
Dendrimers	Hydrophilic/hydrophobic	Medical and biomedical applications.

6. HEALTH AND SAFETY RISKS

In occupational hygiene, one must keep in mind that the direct human health risk caused by an NP first depends on the probability of exposure and, if applicable, on the concentration and duration of exposure. Secondly, it depends on whether these materials, once in the body, exhibit specific behaviour associated with their nanostructure (Oberdorster *et al.*, 2005c).

This chapter thus succintly summarizes the current state of knowledge of the main known health risks of NP. The reader interested by a more detailed review is invited to consult the updated literature review produced by our research team (Ostiguy *et al.*, 2008), as well as Work Safe Australia (2009), Stone *et al.* (2009), Hanai *et al.*, 2009, Kobayashi *et al.*, 2009 and Shinohara *et al.* (2009a). We must mention from the outset that nanotechnology is a rapidly emerging field, that new NP are synthesized regularly and that they often exhibit exceptional physical, chemical and electrical properties.

What about their biological properties and interactions with the human organism? Do they cause a specific risk to the health of the workers who produce, handle, transform or use them? In this new field, there are still major uncertainties and a significant lack of knowledge of the specific health risks of NP. There is still not enough understanding of how NP can be absorbed by the organism, how they will move within it (translocation) or how they will interact with the tissues and biological fluids. On the other hand, Maynard (2006) very clearly shows that, to obtain NP-specific effects, the NP must be capable of interacting with the organism so that the nanostructure is available and so that the material has the potential to induce a biological response associated with its nanostructure.

The existing data coming from exposure studies on cell cultures (*in vitro*), on animals (*in vivo*) or on humans (volunteer or epidemiological studies) and response to respirable particles of nanometric or micrometric dimensions, as well as the general knowledge of a specific product's toxicity, can serve as a basis for preliminary assessment of the risks associated with exposure to a specific NP. In general, it has been shown that at equal mass, the biological response following exposure to insoluble or not very soluble manufactured NP or to ultrafine particles (UFP) of nanometric dimensions is often greater than for the same product at micrometric dimensions, particularly with regard to pulmonary inflammation, tissue damage and lung tumours (Ostiguy *et al.*, 2008). In fact, the biological effect measured for NP seems more correlated to the surface, size and number of particles than to mass, although several other parameters can contribute substantially to the toxic response.

This means that a satisfactory characterization of exposure can currently no longer be limited to product mass, even though the scientific literature cannot yet determine the best parameter to measure exposure. In this sense, Warheit (2009) recommends that toxicologists characterize NP at least in terms of the following parameters: dimensions and granulometric distribution of elementary NP, their shape, specific surface, surface charge, composition and purity, crystalline structure and level of crystallinity, coatings and surface composition, aggregation state, surface reactivity of the particles, and synthesis method, including post-synthesis treatments. ISO (2009a) considers these parameters as essential for the charactherization of NP for toxicological studies: aggregation/agglomeration state, chemical composition, particle size and granulometric distribution, shape, solubility/dispersibility, specific surface, surface chemistry and charge density.

Certain NP and their aggregates may present a serious risk of occupational exposure, likely to lead to pulmonary or cutaneous deposition, with the possibility of translocation in this organism. This is why most OHS concerns pertain to the manufacturing of free NP not bound to materials (NIOSH 2007, 2009b).

This chapter is divided into two sections. The first sets out some of the documented toxic effects in animals or humans related to CNT. The second section is a general discussion, in which the toxic risk is considered in terms of the target organ and the current knowledge of toxicity of ultrafine dust particles (UFP) of aerodynamic dimensions smaller than 100 nm.

6.1 Health Risks Related to Specific Nanoparticles

As a second edition of our literature review on the health risks related to NP has been published (http://www.irsst.qc.ca/files/documents/PubIRSST/R-589.pdf) less than two years ago (Ostiguy *et al.*, 2008), the current section presents only the most recent and significant studies related to the toxicity of CNT, and adding useful additional information.

Contrary to NP, the toxicity of CNT increases with their agglomeration (Swiss Engineering, 2006). These agglomerates resemble asbestos fibres both for their appearance and their toxicity (Swiss Engineering, 2006). From the toxicological point of view, CNT are fibres and their toxicity will be correlated to their biopersistence in the lungs (Lam *et al.*, 2006). However, Muller *et al.* (2006) affirm that CNT inhalation exposure in industrial environments is expected to be very weak, given the propensity of CNT to form agglomerates with an aerodynamic diameter above the respirability threshold (> 5 μm). This does not mean that the respiratory toxicity risk is negligible, especially in cases where the nanotubes undergo some kind of transformation (Muller *et al.*, 2006). In their review of the available data on the toxicity of carbon nanostructures, Panessa-Warren *et al.* (2006) clearly show that the experimental protocols used sometimes have a limited physiological significance due to pulmonary overload or obstruction, or the stress related to instillation in the animal. Nonetheless, CNT seem to have more pulmonary toxicity than carbon black or ultrafine quartz.

At equivalent mass, SWCNT had a greater fibrogenic response than quartz. Granulomas developed at the agglomerate deposition sites, while dispersed CNT instead caused interstitial fibrosis which progressed, even after exposure ended. In a recent study in which CNT were better dispersed, Mercer *et al.* (2008) observed a much stronger fibrogenic response (4 times) than in the study by Shvedova *et al.* (2005), as well as the appearance of fewer granulomas. Also in 2008, Baron *et al.* succeeded in generating single-walled CNT for which Shvedova *et al.* (2008) showed that they had interstitial fibrogenic toxicity by inhalation exposure equivalent to the same kind of toxicity reported by Mercer *et al.* (2008) by aspiration exposure.

Poland *et al.* (2008) exposed rat mesothelial membranes to MWCNT of different lengths, amosite and carbon black. They observed pathological behaviour similar to that obtained with asbestos and directly related to the length of the carbon nanotubes, namely inflammation and granuloma formation. This effect was not observed with short MWCNT. These authors express their fear regarding the possibility of long-term development of mesotheliomas and lung cancers after exposure to such fibres in the work environment. Takagi *et al.* (2008) exposed mice by intraperitoneal injection to MWCNT, crocidolite asbestos and fullerenes. At the end of their study

(25 weeks), they observed a mesothelial response to MWCNT, including moderate to severe peritoneal fibrous adhesion and peritoneal tumours. Crocidolite led to the same effects, but less severely. Finally, fullerene did not lead to these effects.

Tabet *et al.* (2009), using multi-walled CNT on human pulmonary mesothelial and epithelial cells, showed that CNT form agglomerates on the surface of the cells, leading to a decrease in metabolic activity without alteration of the permeability of the cell membrane or apoptosis. They did not observe any internalization or oxidative stress, in contrast with asbestos, which penetrated the cell, reducing metabolic activity without reducing cellular permeability. However, they observed an increase in apoptosis with asbestos.

Tejral *et al.* (2009) reviewed the toxicological impact of CNT. They concluded that in general, functionalized CNT are more toxic than purified CNT, and that many factors affect their toxicity. Among these, it is important to mention the functionalization of the surface, the presence of impurities such as traces of metals, dispersion, deagglomeration and post-purification treatments, which can alter properties such as length, purity, the degree of agglomeration, and the structure. Other properties are just as important, including chemical reactivity, biopersistence, bioavailability, and dimensions. CNT can cause oxidative stress, inflammation, cell damage, harmful effects on cell proliferation, and pathological effects over the long term, such as the formation of granuloma and fibrosis (Tejral *et al.*, 2009). These effects are dose- and time-dependent.

This comparison recalls the problem of asbestos in Québec with an increase in the incidence of pleural mesothelioma cases in the period from 1982 to 1996 (De Guire *et al.*, 2003). In 2008, asbestos remained the substance that caused the most work-related deaths in Québec. Of the 195 deaths recognized by the CSST in 2008, 85 were asbestos-related (CSN, 2009). The studies reported in the current document tend to support the hypothesis that CNT can display toxicity similar to that of asbestos.

Furthermore, Tran *et al.* (2008), in doing a critical analysis of the literature on asbestos and NP with a high length/diameter ratio (High aspect ratio nanoparticles [HARN]), arrive at the conclusion about the fibre paradigm:

- Diameter: fibres must be thin to cross the ciliary region. The aerodynamic diameter of fibres is not substantially affected by their length;
- Length: essential attack parameter, for example of frustrated phagocytosis or other inflammation pathways;
- Biopersistence: important parameter when establishing threshold limit values.

Their review identified many similarities between HARN and asbestos fibres relating to their physicochemical properties and toxic effects. The authors concluded that the evidence suggests that HARN that have the same characteristics (diameter, length and biopersistence) as pathogenic fibres will have similar effects.

While the studies are limited, many toxic effects have already been documented with different NP. In this context, we will see later that several countries recommend that the precautionary principle

be applied to NP with special emphasis on the risks related to CNT, while the EPA treats CNT as a new substance, henceforth refusing to consider them on the same footing as other products with the same chemical composition and with the same standard as graphite. The SCENIHR (2009) also raises concerns regarding the risks of CNT.

6.2 Scopes and Limits of the Current Toxicity Data Related to Nanoparticles

There are many uncertainties in determining whether the unique properties of NP, which are the basis of commercial interest, represent specific occupational health and safety risks. While our previous study (Ostiguy *et al.*, 2008) and the previous section summarized current knowledge relating to the toxicity of specific NP, the fact remains that the latter have been the subject of relatively few studies, mainly concerning the important diversity of synthesized or commercially available NP. Also, the data available for a specific NP are normally only fragmentary, when they exist, and cover only a fraction of the information required to come to a conclusion about its toxicity or harmlessness. Still poorly understood are the factors essential to the prediction of health risks such as the exposure pathways, the translocation of material once it has been absorbed in the body, and the interaction of the NP with biological systems (NIOSH, 2009b).

Soluble NP dissolve and their effects are related to their different components, independent of the particle's initial size. However, the situation is completely different for insoluble or not very soluble biopersistent NP (Oberdörster, 2005a, 2005b). The data currently available on the toxicity of insoluble NP are still limited and still do not allow a quantitative risk assessment (Kuempel, 2006a) or the extrapolation to humans for synthetic NP.

Nevertheless, the data currently available reveal some information that, although preliminary, already allows the conclusion that NP must be handled carefully and that the worker's exposure must be reduced to a minimum because several toxic effects have been documented, with these effects being extremely variable from one product to the next. Along these lines, the IRSST has published a good practices guide suggesting different approaches allowing safe work with NP (Ostiguy *et al.*, 2009; http://www.irsst.qc.ca/files/documents/PubIRSST/R-586.pdf).

In workplaces, NP will normally be absorbed mainly via the pulmonary route and, in some situations, via the cutaneous route and oral route. Depending on their aerodynamic dimensions, inhaled airborne NP can deposit along the entire respiratory tract and not just in the alveoli. Because of their extremely small size, a fraction of the nanometric particles can reach the extrapulmonary organs. This involves a migration of some solid particles, translocation through the pulmonary epithelial layers to the blood and lymphatic systems, as well as through the nerve endings of the olfactory nerves, along the neuronal axons to the central nervous system (Ostiguy *et al.*, 2008).

Another potentially important exposure pathway for workers is skin exposure. In fact, skin absorption could be a more important exposure pathway than inhalation for workers who handle nanomaterials prepared and used in colloid form (Colvin, 2003b). Lipid soluble NP could possibly move in the intercellular space of the horny layer and cross the cells, hair follicules or sudoriferous ducts (Monteiro-Riviere and Inman, 2006). The non-vascularized region of the skin could also serve as a reservoir for NP, from which they could not be eliminated by the macrophages

(Monteiro-Riviere and Inman, 2006). The NP absorbed by the skin could be found in the systemic circulation after having crossed all the layers of the skin (Monteiro-Riviere and Inman, 2006). Absorption can be made easier if the skin's horny layer is damaged. Also, workplace exposure conditions (for example, the humidity or pressure related to handling) can have an impact on skin absorption. In the case of NP poorly absorbed by the skin, an allergy and/or contact dermatitis could be observed (Monteiro-Riviere and Inman, 2006). Warheit *et al.* (2007) conclude that, in the great majority of situations, potential pulmonary absorption in the workplace would be at least one order of magnitude greater than skin absorption.

Good personal hygiene practices in the workplace should greatly limit the ingestion of NP. Nevertheless, NP can be found in the gastrointestinal tract after swallowing, and NP coming from the respiratory system via the mucociliary elevator can be found in the digestive system. The translocation of NP from the intestine to the blood and lymph is possible for some NP (Zhao *et al.*, 2007).

Following exposure, some insoluble NP may possibly be found in the blood after having overcome the respiratory, skin or gastrointestinal protection mechanisms. From there, they will be distributed towards the different organs, throughout the body, including the brain, and can even be stored inside the cells. Furthermore, Oberdörster *et al.* (2007) clearly showed the scientific consensus relating to the propensity of NP to cross cell barriers. Once inside the cells, the NP interact with the subcellular structures, which leads to the induction of oxidative stress as the main action mechanism of NP. Toxic effects have already been documented at the pulmonary, cardiac, reproductive, renal, skin and cellular levels during studies on laboratory animals. Significant accumulations have been shown in the lungs, brain, liver, spleen and bones.

Another difficulty consists of determining the parameter that best links the exposure to the observed effects. In fact, for most chemical products, a good correlation is observed between the exposure dose expressed as mass of product and the measured effects. In the case of NP, countless studies have demonstrated that the mass is not a unit for obtaining a good correlation with the observed effects. In fact, the main data available suggest majoritarily, from the body of knowledge developed with the rat, that at equal mass, NP are more toxic than microparticles for the same product (Oberdörster *et al.*, 1990, 1992, 1994; Heinrich *et al.*, 1995; Driscoll, 1996; Lison *et al.*, 1997; Tran *et al.*, 2000; Brown *et al.*, 2001; Bermudez *et al.*, 2004; Ostiguy *et al.*, 2006a, 2006b, 2008, 2009; Maynard, 2006). Several studies have demonstrated that at equal mass, the toxicity increases as a function of the surface of the NP. Reported by Ferin *et al.* (1992) working with titanium dioxide NP, this difference in reactivity has been confirmed by several research teams and on different products, mainly metallic cobalt (Zhang *et al.*, 2000), titanium dioxide, TiO_2 (Oberdörster *et al.*, 1992; Heinrich *et al.*, 1995; Borm *et al.*, 2000; Renwick *et al.*, 2004) and carbon black (Li *et al.*, 1996; Gallaher *et al.*, 2003; Gilmour *et al.*, 2004; Renwick *et al.*, 2004).

In fact, many studies conclude that a good dose-response relationship exists, but the dose parameter best correlated with the observed effects varies from one study to the next (Agence nationale de recherche, 2005; Ostiguy *et al.*, 2006a, 2008; Oberdörster *et al.*, 1992, 1994, 1995, 2005a; Lison *et al.*, 1997; Maynard et Kuempel, 2005; Tran *et al.*, 2000; Duffin *et al.*, 2002; Brunner *et al.*, 2006; Warheit 2009). Table 3 presents an overview of these parameters.

Table 3 : Main Parameters Allowing the Proper Characterization of Nanoparticles for Toxicological Studies

Parameters
▪ Mass, concentration ▪ Chemical composition (purities and impurities) ▪ Solubility ▪ Specific surface ▪ Number of particles ▪ Size and grain-size distribution ▪ Surface properties (charge/zeta potential, reactivity, chemical composition, functional groups, Redox potential, potential to generate free radicals, presence of metals, surface covering, etc.) ▪ Shape, porosity ▪ Degree of agglomeration/aggregation ▪ Biopersistence ▪ Crystalline structure ▪ Hydrophilicity/hydrophobicity ▪ Lung deposition site ▪ Age of the particles ▪ Producer, process and source of material used

In a context where the number of parameters that can influence the toxicity of NP is as high as this, it is not very realistic to think that studies on the evaluation of the toxicity will be preceded by as exhaustive a characterization of the NP examined. This is why Oberdörster (2005c) and Maynard (2006) recommended several years ago that at least the following physical parameters be evaluated: the number of particles and their dimension (grain-size distribution), the specific surface and the mass. The discussions of more than 70 experts gathered at an international workshop (Warheit *et al.*, 2007) led to the conclusion that it is mainly dissolution, the specific surface and surface characteristics, the dimensions and grain-size distribution, and the shape that essentially determine the functional, toxicological and environmental impacts of NP. More recently, Warheit (2009) recommended that toxicologists characterize NP at least according to the following parameters: the dimensions and grain-size distribution of elementary NP, their shape, specific surface, surface charge, composition and purity, the crystalline structure and degree of crystallinity, the coatings and the surface composition, the state of aggregation, the surface reactivity of the particles, and the synthesis method including post-synthesis treatments. In a recent study, Kroll *et al.* (2009) concluded that the current approaches to toxicological tests have many limitations because of the specific properties of NP. Several of the properties of NP, such as the large absorption capacity, the hydrophobic character/hydrophilic character, the surface charge, the optical and magnetic properties, and the catalytic activity can interfere, among others, with the detection systems. This situation requires validation of the systems used, a detailed characterization of the NP, the use of reference materials, and an extensive characterization of the applicability of the test methods used. The challenge of evaluating the toxicity of NP is therefore based on the development of new standardized *in vitro* tests that are not affected by the properties of NP.

In order to have all of the data that would possibly allow an appropriate dose-response relationship to be determined, SCENIHR (2009) and OCDE (2008) recommended that the following characteristics be measured:

Physical properties:	Dimension, shape, specific surface, length/width ratio, state of aggregation/agglomeration, grain-size distribution, morphology, and surface topography, structure including the crystallinity and structural defects as well as solubility;
Chemical properties:	The structural formula and molecular structure, composition including the degree of purity, surface chemistry (composition, charge, tension, reactive sites, physical structure, photocatalytic properties and zeta potential), known impurities as well as additives, phase, hydrophilicity/lipophilicity.

An important limitation of the actual toxicological data lies in the quantity of information available, the complexity of the NP-body interactions, and the difficulty arriving at a definitive conclusion about the toxicity of a specific NP. The two main limiting factors seem to us to be related to the very limited number of studies on the same products using methodologies and representative workplace exposure routes, or related to their use by humans and the often incomplete characterizations of NP making it impossible to ensure that the NP used are identical. This is how our knowledge of the risks related to products with the same chemical composition but of larger size, as well as the risks related to ultrafine particles can be used to advantage and serve as a starting point in a risk analysis process.

6.2.1 Pulmonary Deposition of Ultrafine Dust Particles

As already mentioned, a worker exposed to substances that may be airborne has three potential absorption routes: pulmonary absorption, cutaneous absorption and absorption by ingestion. The lungs are the main route for dust to enter the human body. It is important to understand two complementary aspects. First, only a fraction of the inhaled dusts will successfully enter the respiratory system. Then, only a certain proportion of this fraction will be deposited and retained by the respiratory system, while another portion can be exhaled, without having been retained in the respiratory system. As a result, only the deposited dusts can have a toxic effect, which will be a function of the deposition site. So what is the specific situation for NP?

In the first place, it is appropriate to examine the situation for a wide range of particle sizes. Figure 4 illustrates the internationally recognized standard curves for airway dust of unit density penetrating the pulmonary airways (ICRP 1994; Maynard and Kuempel, 2005; Vincent 2005). Aspiration by the respiratory system of airborne dust depends on a certain number of variables, including particle size, air displacement rate and pulmonary ventilation rate. For air displacement rates of a few metres per second or less, the probability of a particle penetrating the organism by the mouth or nose (inhalable dust) is nearly 100% for particles with an aerodynamic diameter of a few micrometres (μm) or less and decreases to nearly 50% for particles around 100 μm. Particles smaller than 10 μm have a greater than 50% probability of passing through the thoracic zone (thoracic dust) while particles smaller than 10 μm have a definite probability of reaching the pulmonary alveoli where gaseous exchange occurs (respirable particles). This probability is around 50% for particles smaller than 4 μm, or 4,000 nm.

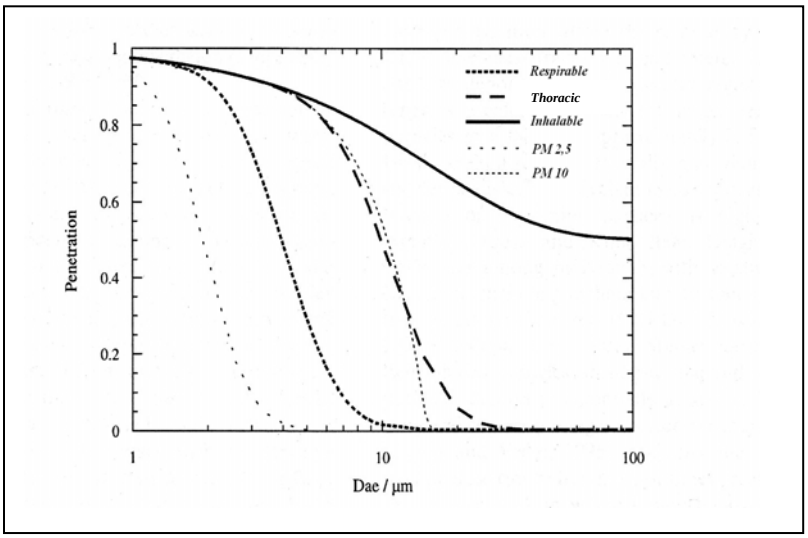


Figure 4 : Standard Curves for Airway Dust Penetrating the Pulmonary Airways

On the other hand, this theoretical curve is not necessarily the same for everyone. Indeed, several factors can alter the structure and functioning of the airway, including workload, sex, age, smoking and respiratory diseases, with consequences both for dust deposition and particle clearance. The model presented here is based on the parameters of a healthy reference population performing an equivalent workload, in a sitting position one third of the time, and executing light tasks for the other two thirds.

As mentioned above, after penetrating the respiratory system, a fraction of the dust will be deposited there and another fraction will not be retained and will be expelled by the lungs. Inhaled dust deposition depends on four main mechanisms, which act differently depending on the particle diameters (ICRP 1994; Maynard and Kuempel, 2005):

- Diffusion is due to random movement (Brownian motion) and has a very strong influence on small particles, causing them to hit a wall, given that they move at random but faster than the air current. This mechanism is more important for smaller particles circulating in an air current at low speed;
- Sedimentation acts on the particle by gravity when the particle can no longer remain suspended in the air current because it is too heavy to be supported by an air current speed that has become too slow;
- Interception occurs when a particle passes near a wall, at a distance of approximately its diameter. It touches an airway and clings to it, resulting in its deposition;
- Inertial impaction occurs when the mass of the particle becomes too great to follow the sudden variation of air flow. It then collides with a wall. This is the predominant mechanism for the largest particles.

Figure 5, taken from Witschger and Fabriès (2005a) and reproduced with the authorization of the Institut National de Recherche Scientifique (INRS) in France, illustrates the deposition rate in the

different pulmonary regions by particle size. However, it should be realized that individual NP can agglomerate in the air and that the pulmonary deposition site will depend on the aerodynamic diameter of the agglomerated particle. This figure clearly illustrates that no particle with 1 nm of aerodynamic diameter, or 0.001 micrometre, reaches the alveoli, while 80% are deposited in the nose and pharynx, with the other 20% located in the tracheal and bronchial region. At this size, retention of inhaled NP is nearly 100%.

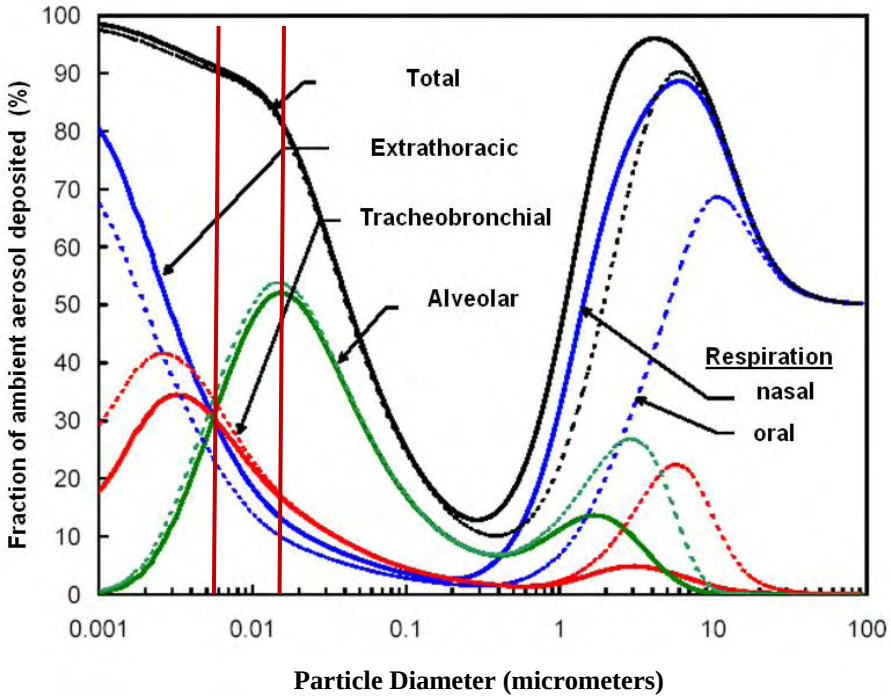


Figure 5 : Prediction of Total and Regional Deposition of Particles in the Airway, by Particle Size

By increasing the particle size to 5 nm (vertical line on the far left), 90% of all inhaled particles are retained in the lung and then are deposited relatively uniformly in the three regions. The total absorption of 20 nm particles (second vertical line to the right) decreases to no more than 80% but more than 50% of the 15-20 nm particles are deposited in the alveoli. This means that 20% of the inhaled particles penetrate the lung but leave it upon exhaling. The particle granulometry thus has a major impact on the pulmonary deposition site and on the quantity of dust deposited in the lungs (ICRP, 1994; Witschger and Fabriès 2005a; Oberdorster 2005b). The deposited dust fraction is greater during exercise (Jaques and Kim, 2000; Daigle *et al.*, 2003) and among individuals suffering from asthma or chronic obstructive pulmonary diseases (Brown *et al.*, 2002b, Chalupa *et al.*, 2004).

Given the major surface differences of each of the three pulmonary regions, even if the mass of 20 nm ultrafine particles deposited in the alveolar region represents more than 50% of the total, the dust concentration deposited per surface unit in the lung will be more than 100 times greater in the nasal region and more than 10 times greater in the tracheobronchial region (Oberdörster, 2005b).

Particle deposition in the extra-thoracic and tracheobronchial regions can play an important role in the development of certain lung diseases, including asthma and chronic obstructive bronchopneumopathies (ISO, 2007). These differences in dust distribution in the lungs may have major consequences on the health effects of inhaled ultrafine particles and on the elimination mechanisms involved (Schiller *et al*., 1988; Kim and Jaques, 2000; Jacques and Kim, 2000; Daigle *et al*., 2003; Oberdörster, 2005a, 2005b; Zhang *et al*., 2005b).

6.2.2 Elimination of Dusts Deposited in the Lungs

The human body has different defence mechanisms for eliminating these undesirable foreign objects. Various processes are involved (Ostiguy *et al.*, 2006a, 2008), which can be divided into two main categories: chemical elimination and physical elimination. The AFSSET (2006) has produced a synthetic table in this sense, which we reproduce here, adapted to our literature review.

Table 4 : Elimination of Dusts Deposited in the Lungs (Synthetic Table)

Nasal region		Bronchotracheal region	Alveolar region
Physical elimination			
Mucociliary movement	X	X	
Phagocytosis by macrophages		X	X
Epithelial endocytosis	X	X	X
Interstitial translocation		X	X
Lymphatic drainage		X	X
Blood circulation	X	X	X
Sensitive neurons	X		
Chemical elimination			
Dissolution	X	X	X
Leaching	X	X	X
Protein fixation	X	X	X

NP and UFP can be eliminated chemically by three major mechanisms: total dissolution, partial dissolution (leaching) or protein fixation. By total dissolution the effects become equivalent to those of larger particles, which are also soluble. Soluble NP and UFP will act on the solubilization site and will not be discussed here, since the effects are highly variable and normally well documented depending on the dust composition. Leaching allows partial solubilization of the particle and part of the effects observed may come specifically from this soluble part. For example, this could be the case for effects observed for CNT associated with the presence of certain partially soluble metals. Finally, other NP or UFP may become fixated to proteins.

The largest particles, which deposit mostly in the upper parts of the lungs, mainly in the trachea and bronchi, are eliminated by different mechanisms including the mucociliary elevator

mechanism where the mucous membranes of the trachea and bronchi are covered with ciliated epithelial cells.

They form an elevator and repel the mucus containing the particles to the digestive system. Normally this is an efficient mechanism that eliminates particles, even ultrafine particles, in less than 24 hours (Kreyling *et al.*, 2002).

At the alveolar level, macrophages will take charge of insoluble particles with a phagocytosis mechanism, whereby the macrophages will surround the particles, digest them if they can and direct them solely to the mucociliary elevator for elimination. This process is relatively slow and has a half-life of over 100 days in humans (HSE 2004a, 2004b; Oberdörster, 2005b). However, the efficiency of phagocytosis largely depends on the particle's shape, number and size. If there are too many particles, the mechanism may become saturated. Several studies seem to show that the unagglomerated ultrafine particles deposited in the alveoli are not phagocyted efficiently by the macrophages (Renwick *et al.*, 2001, 2004). However, macrophages are very efficient in the one to three micrometre range, and thus for much coarser particles (Tabata and Ikada, 1988, Green *et al*., 1998). Pulmonary macrophages cannot efficiently take over fibres longer than 20 micrometres. Once they are deposited, insoluble NP normally can remain in the lungs longer than bigger particles because of reduced clearance and greater retention of particles. Table 4 summarizes the different clearance mechanisms in relation to the pulmonary regions.

The sometimes inefficient takeover of ultrafine dusts by macrophages can lead to a substantial accumulation of particles if exposure continues and to a greater interaction of these particles with the alveolar epithelial cells and bronchi. Studies have shown that some ultrafine particles can pass through the epithelium (epithelial endocytosis) and reach the tissues by interstitial translocation (Oberdörster *et al*., 1992, 2000; Kreyling and Scheuch, 2000). This phenomenon seems to be more significant for higher species, such as dogs and monkeys, than for rodents (Nikula *et al*., 1997; Kreyling and Scheuch, 2000). Once the particles pass through the epithelium, a fraction can reach the lymphatic nodules (lymphatic drainage) or blood vessels (blood circulation) by interstitial transport (Ferin *et al.*, 1992; Nemmar *et al.*, 2002a). Translocation in the human body, meaning transport from one location to another, for NP particles that are insoluble or only slightly soluble, was demonstrated by means of radiomarked carbon NP (Nemmar *et al.*, 2002a, 2002c) while iridium NP show that only a small fraction of the particles are moving (Kreyling *et al.*, 2002).

Two other mechanisms are now recognized for ultrafine particles of nanometric dimensions (Oberdörster, 2005a, 2005b). Ultrafine particles can penetrate the extrapulmonary organs via the bloodstream. Moreover, certain particles can be transported along the axons of the sensitive nerves to the central nervous system. These two mechanisms could play a major role in the development of certain cardiac or central nervous system diseases, but these phenomena have yet to be clearly demonstrated in humans. Katz *et al*. (1984) described the neuronal transport of 20 to 200 nm microspheres from the nose to the brain. The inhalation of 35 nm radiomarked carbon particles led to a significant accumulation in the olfactory bulb of rats seven days after exposure. Several studies showed that when rats are exposed to dusts or welding fumes containing manganese, a manganese fraction could cross the hematoencephalic barrier by circulating directly from the nose to the brain via the olfactory nerves, thus allowing manganese to accumulate in the brain. Such studies were also performed on various insoluble metals and led to the same conclusions (Tjalve and Henriksson, 1999; Brenneman *et al*., 2000; Dorman *et al*., 2002; HSE 2004a, 2004b;

Oberdörster *et al*., 2004a; Ostiguy *et al*., 2003, 2005; Salehi, 2005). Nonetheless, the sensitive nerve translocation studies have mainly been performed in rats. The olfactory mucous membrane occupies 50% of the total nasal surface in rats compared to only 5% in humans. The relative importance of axonal translocation in relation to total absorption has not yet been established clearly in humans. Hankin *et al.* (2008) clearly mention the lack of current knowledge and the research needs regarding the different factors that influence translocation, particularly in the pulmonary interstitium, other lung cells, the blood, the blood vessel walls, the placenta, the fœtus and the brain.

6.2.3 Effects of Inhaled Fine Dusts and Ultrafine Dusts

When particles are reactive or present in sufficient quantity, they can activate or destroy the macrophages or the epithelium and produce an inflammatory mechanism that is pathogenic to pulmonary function. In the event of high and repeated pulmonary dust exposure, the natural defence mechanisms may no longer be enough to do the job. In their model developed in 2003, Faux *et al*. explain how an overload of low solubility dusts can generate oxidizing free radicals, lead to an antioxidant deficit, create oxidative stress and produce inflammation. A whole series of reactions likely to lead to the development of occupational lung diseases are then triggered. Noël and Truchon (2009), in their literature review, also conclude that the interaction between biological material and UFP generates reactive species of oxygen and intercellular stress, which can lead to different repercussions.

Since knowledge of pulmonary toxicity related to nanometric particles is limited, we will report the principal known toxic effects related to respirable or ultrafine particles of up to a few micrometres, i.e. up to over 1000 nm. We will then establish the knowledge level relating to nanometric particles so that we can draw general conclusions.

Dusts found in work environments, often around one micrometre in size, can accumulate in the lungs and lead to several occupational lung diseases, such as pneumoconiosis (asbestosis, silicosis…), smelter's fever, occupational asthma, berylliosis and lung cancer. Donaldson (2005a, 2005b) presents a review of the current knowledge in the field. On the pulmonary level, it clearly appears that toxicity is related to oxidative stress caused by the presence of transition metals, an organic fraction or deposited dusts with a very high specific surface. This oxidative stress can lead to inflammation of epithelial cells.

The most common lung diseases are pneumoconioses, which can be caused by fibrous or non-fibrous dusts and constitute an alteration of the pulmonary structure, resulting from dust accumulation in the lung. This excludes asthma, bronchitis and emphysema (Faux *et al*., 2003). Pneumoconiosis can vary greatly in severity, ranging from very mild to very severe. In mild cases, dust accumulation in the lungs only causes benign effects to the lung structure without harmful consequences. This is often the case, for example, with exposure to iron or tin dusts, which eventually can lead to siderosis or stannosis (HSE, 2004a, 2004b, 2004c). In more severe cases, as often occur following exposure to asbestos (asbestosis) and silica (silicosis), fibrotic changes in the lung lead to major deficiencies in terms of gaseous exchanges. Lung capacity then is greatly reduced and the disease can be fatal. In some cases, these substances, silica and asbestos, have led to the development of lung cancers. Exposure to asbestos fibres can also lead to the development of mesothelioma (HSE, 2004a, 2004b).

Dusts or aerosols reaching the lungs can also cause occupational asthma. In the case of asthma, the lung becomes hypersensitive and subject to constriction, which obstructs the airway. The reaction is often allergic. Bronchitis is a bronchial inflammation, which can also be due to dust accumulation in the bronchial region. This condition can also obstruct the airway, and bronchitis is characterized by a major secretion of mucus (HSE, 2004a, 2004b). Emphysema, often related to cigarette smoking, but which can also result from dust exposure in the work environment, is characterized by the breaching of certain alveolar walls. The direct consequence is that gaseous exchange becomes more difficult, since the available surface for these exchanges is diminished.

Oberdörster *et al*., (1992) showed a translocation of nanoparticles to the pulmonary interstice. This phenomenon was greater with TiO_2 nanoparticles compared to carbon black. The authors associated this difference with the relative facility of deagglomeration of nanoparticles in the lungs. As already mentioned, surface treatments can totally alter the toxicity of nanoparticles, regardless of whether they are found in the lung in the form of unitary particles or agglomerates. Oberdörster (2001) also showed that surface treatment of TiO_2 nanoparticles substantially altered their toxicity. Several pharmacological studies aimed at the development of new drugs exploit this characteristic.

As described in the section on elimination of dust deposited in the lungs, certain nanoparticles can pass through the pulmonary barrier, become systemically available and cause effects elsewhere in the body. The toxicity of these nanoparticles circulating in various organs is not totally known (HSE, 2004a, 2004b, 2004c; Donaldson, 2005a, 2005b; Oberdörster 2005a, 2005b). Nonetheless, animal studies of ultrafine particles have shown pulmonary inflammation with pathological change and translocation of particles to extrapulmonary tissues.

Several studies suggest possible systemic effects of nanoparticles and ultrafine particles (HSE, 2004a, 2004b, 2004c; EPAQS, 2001; Ibald-Mulli *et al*., 2002; Donaldson 1998, 2000, 2005a, 2005b, 2009). According to some authors, the fine fractions of polluting particles, those in the nanometric field, would pass directly from the lung into the bloodstream, alter blood viscosity and thus be responsible for the effects observed (EPAQS 2001; MacNee *et al*., 2000).

Other studies suggest that metal fume fever appearing after acute exposure would also be related to nanometric particles produced during smelting and welding operations and containing different metals, such as zinc or cadmium (Oberdörster *et al*., 1995). The translocation of inhaled ultrafine particles into the bloodstream could affect the endothelial function and promote thrombosis and other circulatory system problems, including increased blood coagulation (Elder *et al*., 2000, 2002, 2004; Kreyling *et al*., 2002; Nemmar *et al*., 2002a; Zhou *et al*., 2003a, 2003b). This phenomenon has been shown in hamsters (Nemmar *et al*., 2002b, 2003) but the situation in humans remains ambiguous.

6.2.4 Epidemiological Studies

Many epidemiological studies have shown a representative statistical link between UFP exposure and different health effects. For example, in a literature review pertaining to diesel engine emissions, Ostiguy *et al.* (1998) had identified several epidemiological studies showing a relationship between exposure to diesel emissions and different effects on exposed workers: cardiovascular symptoms, neuropsychological symptoms, mutagenic effects, bladder cancers and

lung cancers. More recently, Steenland *et al.* (1998), Garshick *et al.* (2004 et 2006), Hart *et al.* (2006) have also found a link between exposure to diesel fume emissions and lung diseases, including cancer and neurological effects in workers exposed to diesel UFP. Many studies also showed links between exposure to welding fumes containing a large proportion of UFP or asbestos and lung diseases.

On the cardiovascular level, in human beings, at the cardiovascular level, epidemiological studies and studies of volunteers have shown that the level of inhaled particles has direct effects on cardiovascular physiology, with alterations of health rhythm and arterial diameter. Moreover, several epidemiological studies (Peters *et al*., 1997; Wichmann *et al*., 2000; Penntinen *et al*., 2001, Pekkamen *et al*., 2002; Mills *et al.*, 2009) identified a direct relationship between exposure to ultrafine dusts of nanometric dimensions and respiratory and cardiovascular effects. Several epidemiological studies have established significant relationships and shown that an increase in fine particle air pollution, originating mainly from vehicle emissions, led to a rise in morbility and mortality of more fragile populations with respiratory and cardiac problems (Pope *et al.*, 2002, 2004; Schwartz *et al.*, 1991, 1993, 1995, 1996; Dockery *et al.*, 1993; Romieu 1996; Pekkanen, 1997; Penttinen *et al.*, 2001; Timonen *et al.*, 2004; Bruske-Hohlfeld *et al.*, 2005; Ruckerl *et al.*, 2006). Controlled clinical studies in the laboratory have shown deposition of ultrafine dusts throughout the pulmonary tree, accompanied by cardiovascular problems (Brown *et al.*, 2002a, 2002b; Daigle *et al.*, 2003; Oberdörster, 2005a, 2005b, Frampton *et al.*, 2006). Studies of coal miners exposed to ultrafine dusts have proved the accumulation of such dusts in the liver and spleen (Donaldson, 2005b). The accumulation was higher in miners displaying severe pulmonary problems, thus suggesting that damaged lungs or lungs with major deposits favour the passage of ultrafine particles into the circulatory system.

To summarize, ultrafine dusts of the same dimensions as nanoparticles enter the body mainly by inhalation and are deposited in the lungs; some of them can reach the brain directly via the olfactory nerves. The lungs do not necessarily succeed in eliminating these undesirable particles, which then cause pulmonary inflammation, potentially leading to the development of certain lung diseases. Moreover, some of these very fine dusts can pass through the various protective barriers of the lungs, reaching the circulatory system; they are then distributed to all parts of the body, where they can cause different kinds of damage. It is not yet very well known how the chemical and physical properties of NP influence translocation. Because of their small size, some NP pass through the cell membranes and interact with the cell structures, such as mitochondria. Oberdörster (2005b, 2007) summarizes the effects on the body of inhalation of ultrafine dusts of nanometric dimensions (Figure 6, reproduced with the authorization of Dr Gunter Oberdörster, (2005b)). The translocated particles can them become mediators, causing various effects on the body.

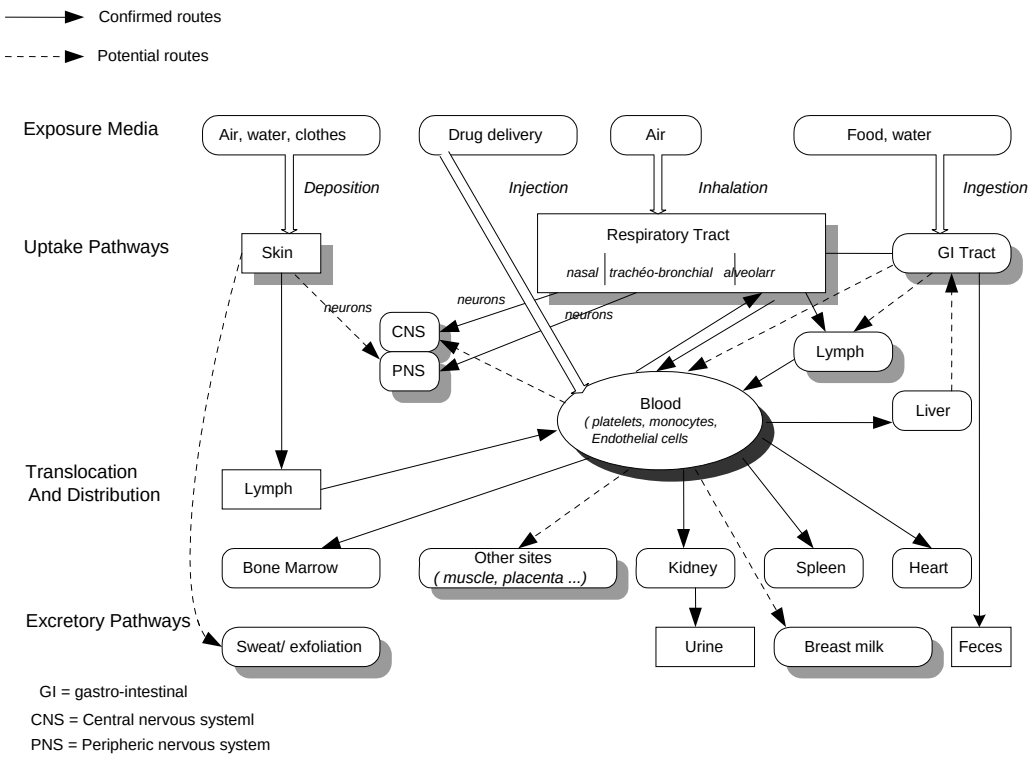


Figure 6 : Potential Effects of Inhaled Ultrafine Particles

6.2.5 Discussion on the Health Risks

Despite the limited information on the health risks related to engineered NP, plenty of knowledge exists on the toxicity of the risks related to UFP of nanometric dimensions. Thus, the biological pulmonary response mechanisms, such as oxidative stress, inflammation, production of chimiokins and cytokins and the cellular growth factors seem to constitute a common response to the two types of NP (Knaapen *et al.*, 2004; Oberdorster *et al.*, 2005a; Donaldson *et al.*, 2005a, 2005b). The pulmonary effects related to the presence of metals also appear to be similar in both cases (Kagan *et al.*, 2006). These similarities make it possible to hypothesize that even if the compositions and the physicochemical characteristics of NP and UFP can be substantially different, the knowledge resulting from the UFP studies could be extrapolated to the synthesized NP in a first approximation.

The CNT studies have shown pulmonary inflammation and fibrogenic reactions formation of granulomas and mesothelial tumors in animals, suggesting potential behaviour similar to asbestos, in which the effects are mainly related to the length (NTC longer and more toxic), the diameter and the biopersistence of the fibres (Lam *et al.*, 2004, 2006; Muller *et al.*, 2005, 2006; Shvedova *et al.*, 2005, 2008; Poland *et al.*, 2008; Takagi *et al.*, 2008; Donaldson *et al.*, 2009; Castranova, 2009). Moreover, the agglomerates generated granulomas, while the dispersed structures produced progressive interstitial fibrosis (Mercer *et al.*, 2008; Shvedova *et al.*, 2008; Poland *et al.*, 2008).

During the INNO-09 conference, Geraci (2009) mentioned that Hubbs *et al.* reported, at the 2009 Conference of the U.S. Society of Toxicology, that MWCNT can penetrate the pleura and cause persistent inflammation and fibrosis. Lindberg *et al.* (2009) demonstrated dose-dependent genotoxic effects following *in vitro* exposures of bronchial epithelial cells to CNT.

However, it must not be forgotten that any surface alteration is likely to alter toxicity substantially. Cytotoxic studies *in vitro* of quantum dots also showed that the surface coating significantly altered cellular viability (Shioara *et al.,* 2004; Lovric *et al*, 2005; Hoshino *et al.,* 2004).

Translocation is another NP property that must be taken into account, because some NP will pass through the organism's different protective barriers. Thus, captured at the nasal level, they may follow the olfactory nerves and penetrate directly to the brain. Several studies have also shown that NP have the property of infiltrating the pulmonary epithelium much faster than coarse particles, and passing into the nearest lymphatic and blood circuits, from which they can circulate throughout the body and be retained by certain target organs. They can also penetrate the different tissues and the cells. This invasion by NP can generate acute inflammatory responses, resulting in the release of certain chimiokins, cytokines and fibrogens. Moreover, the activation of macrophages and neutrophils is associated with the production of active oxygen species, which will be destroyed by antioxidants. If the mechanism becomes overloaded, it can also trigger a persistent inflammatory response, leading to the development of diseases.

Many epidemiological studies have shown a strong link between exposure to particles of nanometric dimensions and the development of occupational diseases. Oberdorster (2007, Figure 6) provided a good summary of the known and supposed potential impacts of such exposures. Although the scientific and medical evidence is insufficient to recommend specific medical detection in workers potentially exposed to NP, this in no way prevents medical surveillance (NIOSH, 2009a) by employers interested in implementing a precautionary approach. If NP have the same chemical composition as a bulk product for which specific medical surveilance exists, it should then be applied to the NP.

6.3 Safety Risks Linked to Nanoparticles

Another risk linked to NP is the occurrence of catalytic reactions or violent reactions, which can be very dangerous (Pritchard, 2004; Biswas and Wu, 2005; Lamy, 2005). Catalytic reactions depend on the composition and structure of the NP (NIOSH, 2006, 2009b). For example, NP and porous materials of nanometric dimensions have been used for years as a catalyst to increase the speed of reactions or reduce the temperature necessary for reactions in liquids or gases (NIOSH, 2006, 2009b).

Pritchard (2004) produced a literature review on the explosion risks associated with NP and found that this aspect has not been documented in the literature to date. We must therefore extrapolate based on knowledge related to fine and ultrafine powders. He points out that this extrapolation of behaviours of larger particles to NP cannot be achieved with certainty, given the major changes of chemical and physical properties of particles smaller than 100 nm. For coarse dusts, the tendency to explosion and the ease of ignition increase as particle size diminishes. Indeed, because of the very fine granulometry, dust reactivity is increased, as are the explosivity parameters. The finer a dust is, the greater the pressure rise and the lower the ignition energy. On the other hand, for some

dusts, this tendency fades and reaches equilibrium when the dust reaches a few tens of micrometres. Normally, the energy required to detonate dust is greater than the energy necessary for gas. NP thus could exhibit a high explosion risk (Pritchard, 2004).

Nonetheless, for the same product mass, combustible NP could present a higher explosion or fire risk than larger particles because of their greater specific surface, the possible decrease in ignition energy and the increase in the combustion rate. Thus, a relatively unreactive particle could become highly combustible when it is encountered at nanometric dimensions. Among the rare data available, Granier and Pantoya (2004) showed that an Al/MoO_3 alloy of nanometric dimensions catches fire more than 300 times faster than the same alloy of micrometric dimensions.

This explosive potential depends on the substances used (type, composition or particle surface characteristics, dimension, etc.), their different nanometric characteristics (solid or liquid state, in a mixture or in bulk) and the quantity of particles produced or present in the air (quantity between the upper and lower explosive limits), and the environmental conditions (oxygen concentration, source of ignition, temperature) (Pritchard, 2004; Proust, 2005). Proust (2005) specifies that dust clouds can explode as long as the particles present are capable of burning in air. He adds that NP-related accidents are considered major because of their high destructive potential. Many scientific projects have been devoted to the study of dust explosions: cloud formation mechanisms, triggering and propagation processes, induced pressures effects, etc. In particular, this has opened the way to accident modelling and the perfecting of risk prevention techniques.

Pritchard (2004) mentions that the maximum size of the particles that can form an explosive dust cloud is 500 µm, while there is no lower limit. Thus, most combustible NP would have the possibility of exploding. The particle surface will accumulate electrostatic charges which, in an NP cloud, can become their own activation energy. In reality, the dust concentration, the composition of the particle, the minimum activation energy (usually between 1 and 10 mJ), the degree of dispersion, the initial conditions (temperature, pressure, particle turbulence), the oxygen concentration, the humidity rate (the higher it is, the less severe the explosions will be) and the solvent rate (increasing the severity), the presence of other substances (altering the particles flammability) and the possibility of forming an airborne particle cloud are all factors capable of influencing the production of an explosion (Pritchard, 2004). The higher the temperature will be, the more the explosion risk will actually increase because the minimum energy will be easier to attain. However the power of the explosion will be reduced (Pritchard, 2004). Two conjugated factors, the low spark ignition limit and the natural production of charges during handling, create a high *a priori* risk that electrostatic sparks will trigger an explosion. This could necessitate the development of specific explosion and firefighting measures (Proust, 2005).

With micrometric particles, the upper explosion limit is often situated around a concentration of 2 to 3 kg/m^3 and the lower limit around 50 to 100 g/m^3. In fact, the particle clouds must be dense enough for the different particles to influence each other or be influenced by an external activation source (Biswas and Wu, 2005; Pritchard, 2004). Closed, cramped or confined areas are especially dangerous for explosions because particles have less space to disperse and thus more chances of having a high concentration and an accumulation of electrostatic charges (Dinyer *et al.*, 2005). The explosion can then occur within pipes or equipment. The available space is a key factor, because a process will have more chance of exploding in a closed vessel or a sealed container than in a large space, because the particle concentration is greater. The accumulation of charges on a particle may

cause calcination of this particle which, in releasing energy in a confined space, will stimulate the same reaction in the neighbouring particles and trigger a chain reaction, which can end in an explosion or a fire.

To summarize, the combined reaction of two or more types of particles and the capacity of substances (or particles) to ignite spontaneously in the air can create an explosion or a fire. The type of process (if there is a risk of ignition) and cleaning (more risk of explosion by vacuuming than by cleaning with water and an inert solvent) are other factors which can lead to an explosion (Dinyer *et al.*, 2005).

The different processes (particularly particle drying), the type of storage (two incompatible substances, one beside the other, can be dangerous, particularly if there are leaks, poor maintenance, containers lacking a tight seal, or even contact by means of the ventilation), the handling (transport, transfer) will influence suspension and formation of particle clouds. A draft above a deposit or a first explosion (elsewhere in the factory) could result in suspension of particles, if ventilation is ineffective. This suspension could trigger a first or second explosion (Pritchard, 2004). The explosion risk with NP can persist over a long period of time if the particle cloud is not dispersed or removed, given that particles smaller than 1 μm have a longer deposition time (Pritchard, 2004). Thus, an explosion is triggered by the formation of a particle cloud that persists for a varying length of time, but long enough to suffer the effects of an ignition source coming from the cloud itself (electrostatic charge, self-warming and pyrophoricity) or from an external source (spark, fire or hot surface) (Biswas and Wu, 2005). Finally, the presence of a fire in the same room or an explosion in another part of the building can trigger an NP explosion, either by a rise in temperature (source of activation) or by suspension of particles triggering the formation of a second explosion (Pritchard, 2004; Kirby, 2005).

Particles of nanometric dimensions have long been used on a large scale as catalysts. Depending on their composition and their structure, some nanomaterials can initiate catalytic reactions and increase the risk of explosion or fire, which normally would not be anticipated based on their chemical composition alone (Pritchard, 2004).

If an explosion occurs, even in the hypothesis of small-scale processes, it is likely to destroy all or part of the process, especially the weakest components, such as the filters. Apart from the direct effects of pressure and eventual projected debris, there is reason to fear dissemination in the atmosphere of a large quantity of NP propelled by the force of the explosion. A toxic dimension is then added, aggravating the overall risk. The explosion hexagon (Figure 7) summarizes the conditions required for an explosion to occur, while Figure 8 summarizes the main situations that can favour such an explosion.

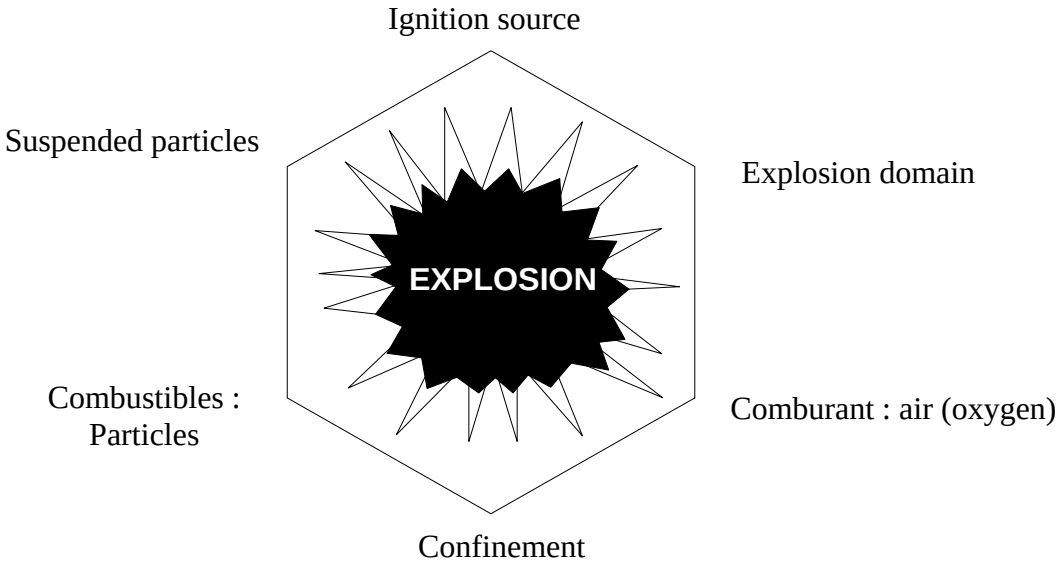


Figure 7: Explosion Hexagon

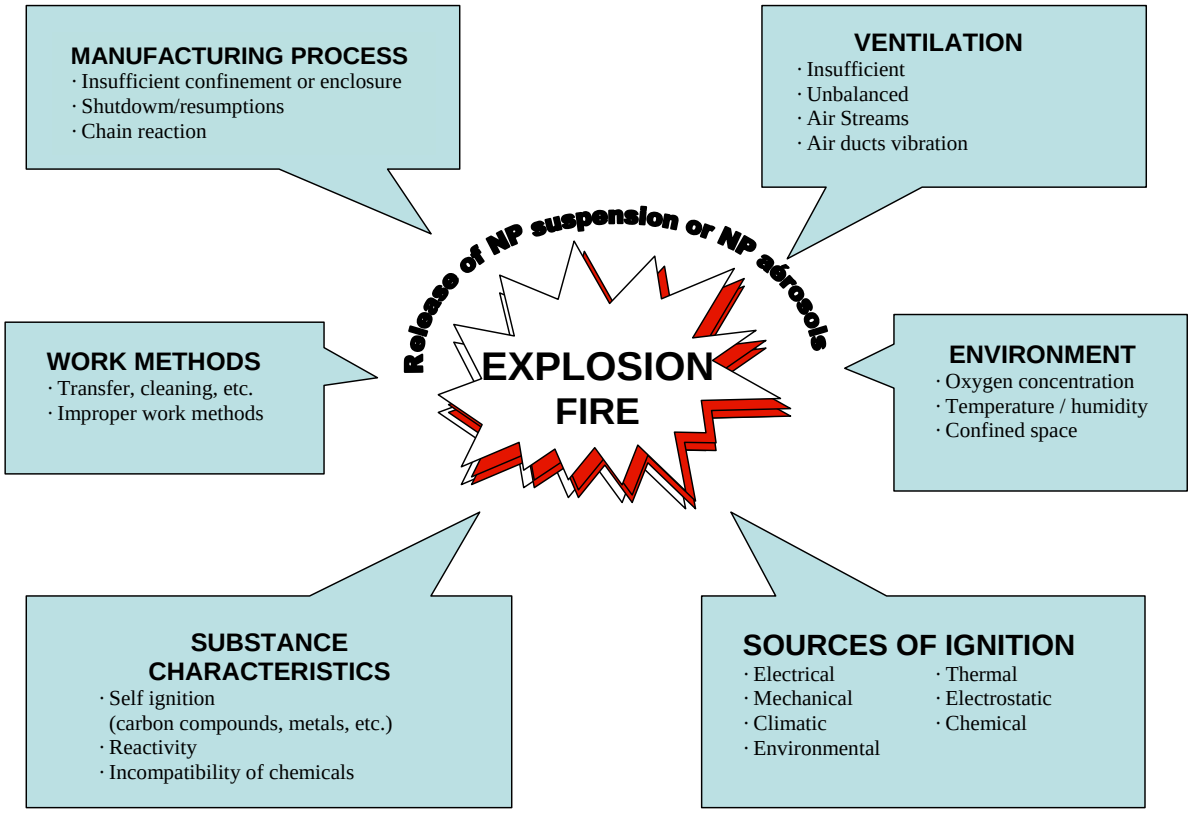


Figure 8: Examples of Conditions Favouring the Occurrence of an Explosion

7. NANOMATERIALS AND THEIR AIRBORNE BEHAVIOUR AND DETECTION

The previous chapter showed that, from now on, the mere measurement of the mass of an NP to which a worker is exposed is no longer a sufficient parameter to assess the toxic risk. However, there is too little information at present to determine the appropriate exposure parameter linking the exposure to the risk and thereby to determine the exposure characterization techniques most suitable for implementation.

In a recent literature review, a group of international experts (Council of Canadian Academies, 2008) concluded that the current scientific knowledge no longer makes it possible to answer fundamental questions, such as what physical properties are most appropriate to measure that allow occupational exposure to be linked to a nanomaterial's biological interactions. Thus, they recommend that multiple measures be applied to characterize a material during exposure: size, mass, composition, surface area, shape and morphology, crystallinity, surface charge, surface chemistry, solubility in lipids or in water, aggregation and agglomeration. They also conclude that, in most cases, the traditional measuring tools are deficient and limit scientific progress.

NP characterization, which may differ from the characterization normally used in occupational hygiene, requires a specific instrumentation, which must measure a minimum number of parameters, including mass, number, size and granulometric distribution of NP and agglomerates, chemical composition and specific surface. The characteristics of NP surface properties would also have to be documented more. Only these additional data will eventually allow NP exposure to be linked to toxic risk. However, given that the techniques necessary for measurement of these toxicity-related characteristics are not available today for sampling at employee workstations, certain instruments will have to be developed. The OECD (2008), the ISO (2009), Warheit (2009) and the SCENIHR (2009) propose the parameters recommended for characterization of engineered NP (see paragraph 6.2).

All of these factors combined go far beyond the objectives of our report. This chapter thus will be limited to summarize the characteristics of different instrumental techniques which can help document some of their specific parameters and contribute in the characterization of NP[3]. The next chapter, complementary to this one, will focus on the development of strategies capable of documenting occupational exposure based on the available instruments.

7.1 Definition

As already mentioned, there is now unanimity in the scientific community on the dimensions of manufactured NP: at least one of their dimensions ranges between one and 100 nm and they have different properties than larger-diameter particles made of the same material (ASTM, 2006; BSI, 2008; ISO, 2007, 2008). Figure 9 allows the aerodynamic diameter of some aerosols to be compared, allowing NP smaller than 100 nm to be positioned relative to other particles.

[3] This aspect was discussed in detail in the first edition of this document. Only a brief summary is presented here. A reader interested in more information is invited to consult Ostiguy *et al.* (2006b).

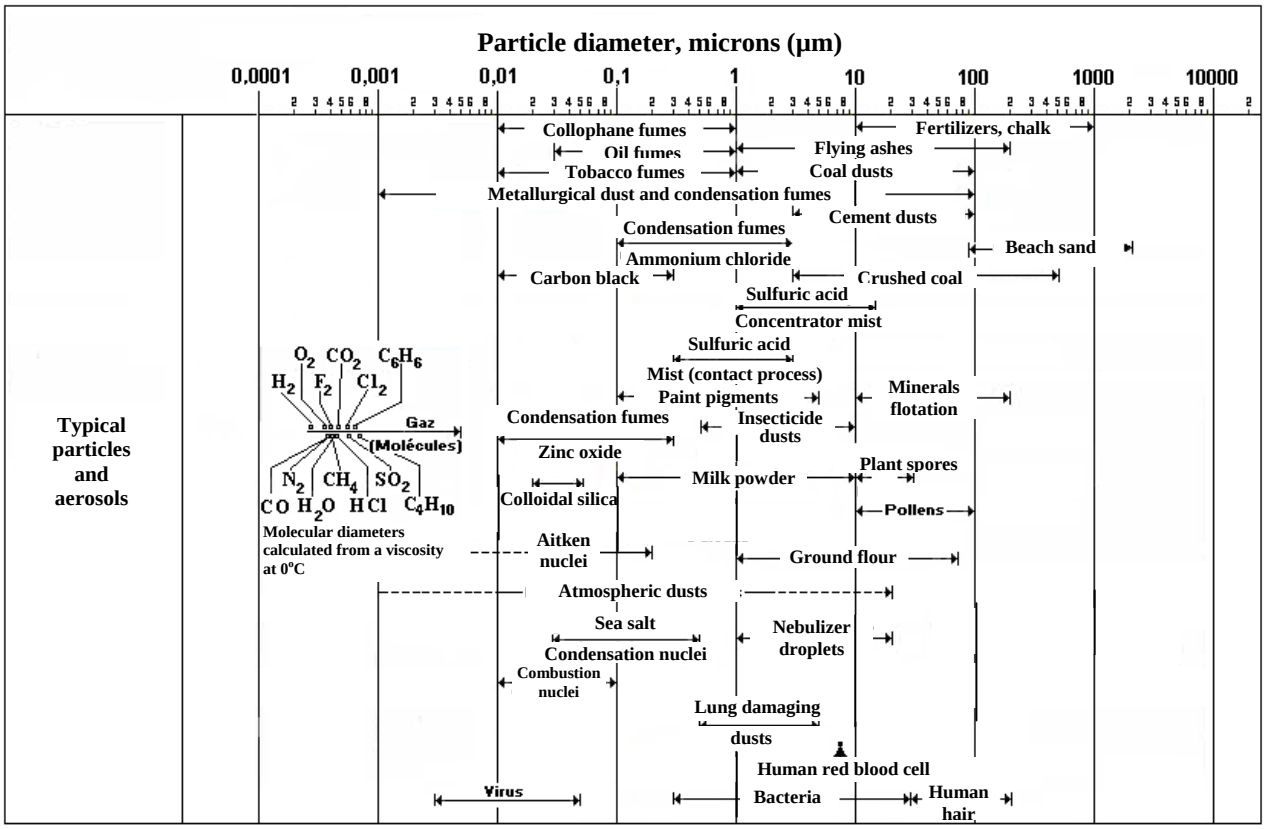


Figure 9 : Distribution of the Diameters of Particles Commonly Found in Workplace Air

7.2 Formation and Behaviour of Nanometric or Ultrafine Particles

We will summarize here the different diffusion, coagulation, aggregation, agglomeration, gravitational sedimentation, as well as resuspension processes involving NP.

Diffusion is normally the principal mode of transport of nanoparticles. Diffusion always occurs from the highest concentration to the lowest. The smaller the particle, the more rapidly it disperses. At 1 nm, a particle's diffusion coefficient is about 5% of the diffusion coefficient of gases (Maynard and Kuempel, 2005). Thus, NP will not remain localized in the zone near a leak in the work environment. They will diffuse rapidly over a great distance and potentially will expose many individuals far from the leak. In many situations, including during inhalation by the worker, the diffusion speed is much greater than that of the air current in which the NP is found, so that the NP will have a very random trajectory (Brownian motion), strongly influenced by its collisions with gas molecules, vapours or other dust particles.

The primary aerosol nanoparticles that come into contact will adhere to each other to form aggregates ou agglomerates. The usual difference between these two terms is the following. In aggregation, the particles bind strongly to each other such that their total surface is less than the sum of the surfaces of the individual particles. Aggregates are difficult to take apart.

Agglomeration normally means that the particles are weakly bound to each other, are relatively easy to separate, and that the total surface area of the agglomerates is identical to the sum of the surfaces of the individual particles comprising them (ISO, 2007). NP have a natural tendency to agglomerate, which is even stronger because the diffusion produced by Brownian movement promotes collision between them. Thus, there is an increase in the aerodynamic diameter of aggregated or agglomerated particles, and thereby a decrease in the number of particles (Maynard and Kuempel, 2005). The speed at which these particles will agglomerate (called the coagulation rate) will depend on their numerical concentration and their mobility, which is inversely proportional to their diameter. Figure 10 shows the relationship between the numerical concentration of airborne particles and the time required for coagulation of monodispersed aerosols.

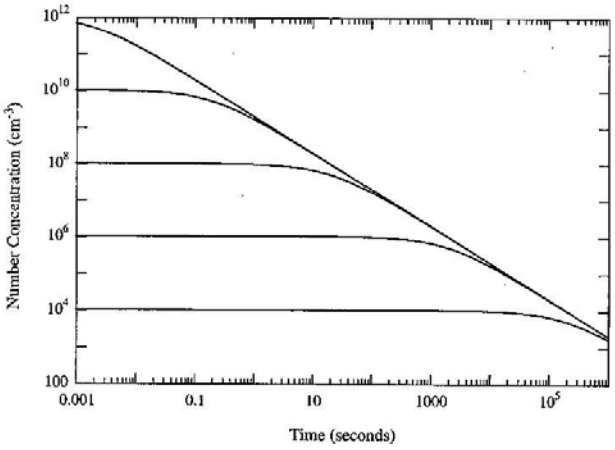


Figure 10 : Relationship between the Numerical Concentration of Particles and the Time Required to Coagulate

An aerosol's primary particles will adhere to each other to form aggregates strongly held together by ionic/covalent bonds and difficult to disperse again by mechanical means (BSI, 2005; NIOSH 2006, 2009b). Preining (1998) showed how the aggregation speed of particles is linked to their size and their airborne concentration. Table 5 clearly illustrates how 10 nm particles may need a few milliseconds for 50% of them to coagulate if their concentration is around 1 g/m³. On the other hand, also for particles of this size, the time required for 50% of them to coagulate will increase to 11 minutes if the concentration is 1 μg/m^3.

Aggregates can bond to each other to form weakly bonded agglomerates. They tend to adhere to each other due to Van der Waals forces, which act only over short distances, and to electrostatic forces present in the particles and the surface tension related to the adsorbed liquids (BSI, 2005; NIOSH 2006).

The coagulation is thermal when caused by Brownian movement and kinetic when caused by an external force such gravity, electrical forces, or aerodynamic effects. On the terminological level (ISO, 2007), the elementary nanoparticle, which has a diameter smaller than 100 nm, can form a larger agglomerate called a "nanostructured particle", while NP aerosols or nanostructured particle aerosols are called "nanoaerosols".

Table 5 : Nanoparticle Coagulation Time as a Function of their Size and Concentration

Particle Diameter (nm)	Half-life Time			
	1 g/m^3	**1 mg/m^3**	**1 µg/m^3**	**1 ng/m^3**
0,5	0,39 µs	0,39 ms	0,39 s	6,5 min
1	2,2 µs	2,20 ms	2,2 s	36,67 min
2	12 µs	12 ms	12 s	3,34 hours
5	0,12 ms	0.12 s	2 min	33,34 hours
10	0,7 ms	0,7 s	11,67 min	8,1 days
20	3,8 ms	3,8 s	63,34 min	43,98 jours

The coagulation of very small particles (including those ranging from 1 to 100 nm) thus will lead to the formation of bigger, less mobile and less numerous particles to attain a state of equilibrium, in which their growth will be much slower. As they reach a size of around 100 nm, they will grow less rapidly up to a size of about 2,000 nm. This slowed growth range, between 100 and 2,000 nm, is called “accumulation mode”.

The larger the dimensions of the particle, the heavier it becomes in relation to air, and the greater effect that gravity will have on its movement. In fact, some particles suspended in the air can be deposited (on the ground, on workers, on equipment, tools, walls, beams, work surfaces). This is the phenomenon of gravitational sedimentation. Under identical air turbulence conditions, the heavier the particle, the faster the settling rate will be.

All of these phenomena lead to the conclusion that the smaller the particles, the longer distances they can travel, with their dimensions slowly increasing and their composition evolving over time through impacts with other particles. They will end up being heavy enough for sedimentation. The time required for the NP to grow in size and the considerable distances that it can travel before settling in no way represents an effective approach for controlling its concentration in the air.

Several processes are used for preparation of NP in the form of powders, which can be resuspended in the air at different stages of the industrial process, such as synthesis, transfer, drying, bagging, unbagging, handling… They can also be resuspended during maintenance or repair operations or during accidents involving an overturned vehicle or a spill. Poor maintenance of the workplace can result in resuspension of deposited dust by drafts or by human or mechanical activities.

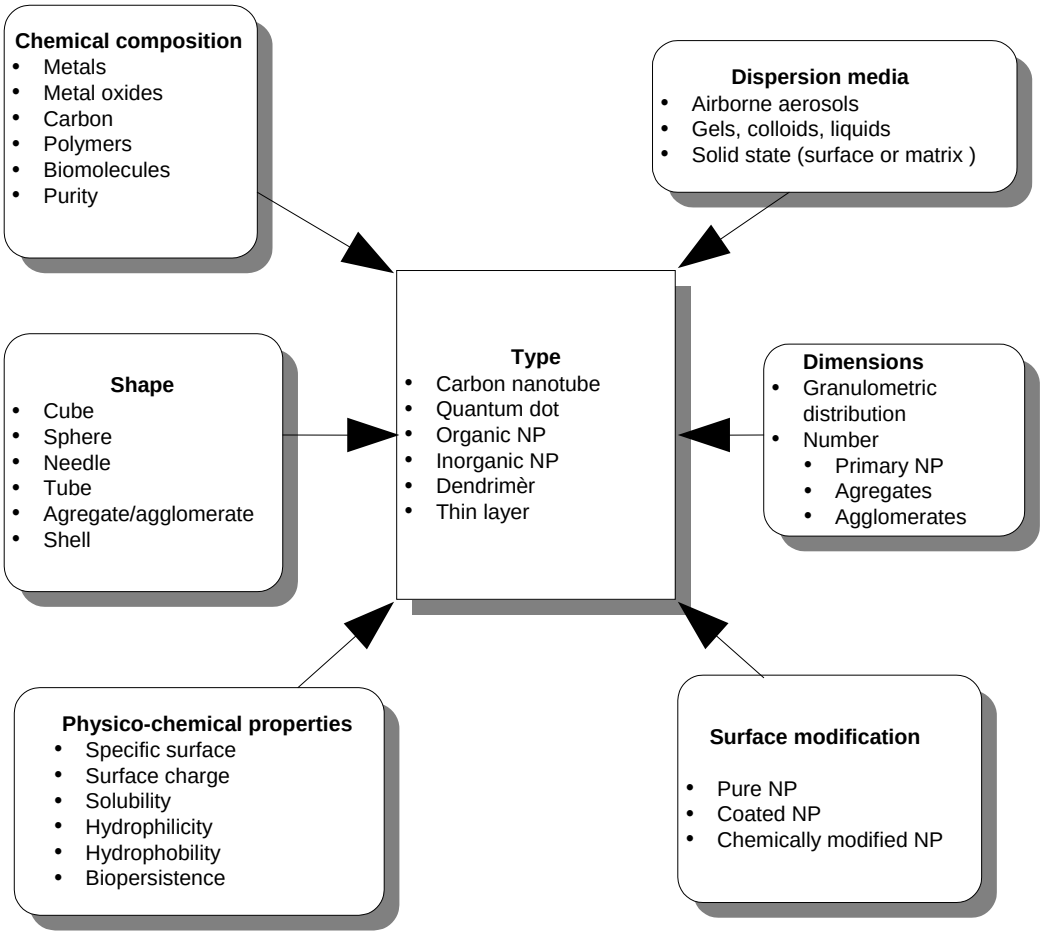
The propensity of particles to resuspend is extremely complex. Several factors specific to NP can influence this, particularly size, shape, electrostatic charge of particles, their surface characteristics and ambient humidity. External energy (mechanical energy, agitation, drafts, etc.) can play a major role in resuspension of dust particles already sedimented. Maynard (2004) (see subsection 8.2) showed that CNT resuspension could prove relatively difficult due to certain properties inherent to the initial products, while it is very difficult to weigh nanometric silica or titanium dioxide because the slightest draft can resuspend these substances in the air very easily.

7.3 Detection of Nanoparticles

The three main absorption routes for workers exposed to NP are inhalation, ingestion and cutaneous absorption. As in the case of other dusts, inhalation normally is the main NP absorption route. The different strategies for assessment of occupational exposure will normally target the inhalation route first.

Different motivations lead occupational hygienists to sample NP in order to protect workers. The quantitative evaluation of exposure and adequate characterization of engineered NP aerosols currently represent a serious challenge. Figure 11 represents certain variables and physicochemical characteristics of NP, which must be taken into account in the risk assessment and which consequently must be specified when assessing occupational exposure (FIOH, 2007; Ostiguy *et al.*, 2008, 2009).

Figure 11 : Variables and Physico-Chemical Characteristics of Nanoparticles Useful for Risk Assessment



Moreover, any environment of the industrial establishments already contains an often complex mixture of UFP in the indoor and outdoor air, regardless of whether these UFP are of natural origin or whether they come from industrial processes. Through this mixture of particles of different granulometries and various compositions, the content of the targeted engineered NP must be determined and they must be characterized.

Different instruments or techniques allow measurement of parameters for particles suspended in a given liquid or gas volume, in general, the particulate concentration in mass, number or surface. These instruments or techniques are also capable of obtaining the granulometric distribution of aerosols, i.e., the measurement of these parameters for the different sizes of particles present in the aerosol. Figure 12 summarily illustrates a few of these techniques and their field of applicability.

With the development of well characterized reference materials in the nanometric size, calibration of instrument will be possible.

Contrary to our first edition (Ostiguy *et al.*, 2006b), we have chosen to present, very briefly, the different types of instruments based on the NP parameter allowing correlation of exposure to the health effects and by referring the reader to this first edition for additional details on the performances of these instruments as well as a partial list of the commercially available equipment. This orientation is similar to that of the ISO technical reports (2007, 2008b), among others, which have the objective of characterizing UFP aerosols, NP aerosols and nanostructured aerosols.

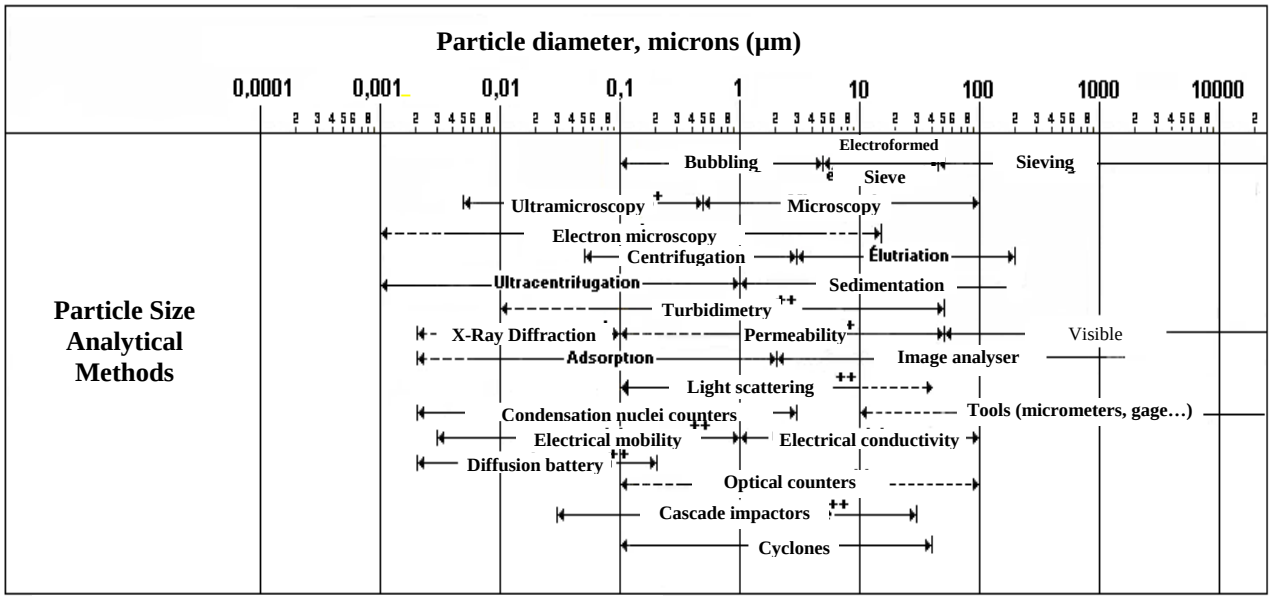


Figure 12 : Some Techniques Usable for Airborne Dust Characterization

Ideally, as in the case of other contaminants, NP exposure should be assessed with portable, practical instruments, easy to use in the work environment while not necessitating very advanced expertise. Among the characteristics sought, in addition to portability, we note the possibility of collection in the breathing zone and measurement of different parameters (number of particles, mass, specific surface, charge, granulometric distribution, degree of aggregation, spatiotemporal variations, etc.) essential to the establishment of the relationship between exposure and the health risk. Moreover, they should be able to discriminate between engineered NP and dust particles of the same size present in the work environments and coming from natural sources (volcanic activity, erosion, marine aerosol, etc.) or anthropic sources (incomplete combustion of fossil fuels, welding fumes, etc.) and be robust, reliable, low-cost and autonomously powered whenever possible. As we will see, such instruments are not yet available. The ISO (2007) standard mentions that no perfectly conforming granulometric selector with a 100 nm cut-off diameter is currently available on the market.

The main NP parameters that can be measured by the instruments we describe here are mass concentration, number of particles, their specific surface and their granulometric distribution.

- Mass concentration

Even though the mass concentration does not represent the best parameter for assessing NP exposure, determining this parameter nonetheless can provide valuable information. In the absence of a selective instrument measuring the parameters of particles with a 100 nm cut-off diameter, several instruments can help document this concentration. Impactors allow separation of aerosol particles into different granulometry brackets. Impactors offer the advantage of allowing gravimetric determinations and subsequent laboratory analysis of particles on the collection substrates. A wide range of impactors exists but some have been developed specifically for ultrafine particles. They have a lower detection limit and can classify particles from 10 nm. Among them, the electrical low pressure impactor (ELPI) simultaneously measures the granulometry and the number of particles in each granulometry range.

Tapered element oscillating microbalances (TEOM) allow the determination of the mass of NP. In a TEOM, the particles are sampled on a collector substrate located on the tip of an oscillating glass tube. The oscillation frequency depends on the mass of dust particles sampled on the collector substrate. These instruments, frequently used continuously to determine environmental dusts, are reliable and show a detection limit of around 0.01 μg for a one-hour collection time. They require a pre-filter carefully chosen to eliminate excessively coarse particles and an efficient collector substrate for quantitative NP filtration. These instruments are widely used in environmental studies for evaluation of PM_{10}, $PM_{2.5}$ and PM_1.

The piezobalance allows direct deposition of dust on a crystal by impaction or electrostatic precipitation. The deposited dust can be measured by determining the crystal resonance frequency. In the case of this balance, the dust deposition efficiency depends on the granulometry and some of the properties of the dust particles, which makes them more difficult to quantify in the work environment. The piezobalance is substantially less sensitive than the oscillating microbalance and the detection limit is around 100 $\mu g/m^3$. On the other hand, the concentrations measured can reach a few percentages and the response time is short, within one minute.

Devices using electrical mobility (DMA, Differential Mobility Analyser) make it possible to classify particles by granulometric sizes. The mass can also be indirectly determined by using a few instruments in tandem. For example, determination of the size distribution, coupled with the determination of the number of particles, would allow an approximation of the mass by knowing the density of the product. SMPS (Scanning Mobility Particle Sizer) and FMPS (Fast Mobility Particle Sizer) are examples of equipment for indirectly determining the mass of particles by size fractions. The particles are classified by means of an electric field, and counted with a condensation nucleus counter (CNC) in the case of an SMPS, and with an electrometer in the case of an FMPS. Cumbersome and hardly portable, these instruments are, however, among the best instruments available for analyzing particles larger than 2-3 nm (ISO 2008a).

- Numerical concentration

Numerical concentration of particles is another essential parameter to measure to characterize NP. Condensation nuclei counters (CNC) count particles. However, this detection is not based on particle size. Serially coupled to a granulometric preselector such as the Differential Mobility Analyser or a diffusion battery (Ostiguy *et al.*, 2006b), CNC can provide real time measurement and detect particles as small as 2-3 nm, give information on the number of particles and the particle size distribution. An electrometer can also be used (Ostiguy *et al.*, 2006b). Finally, electron microscopy can accumulate information, during a subsequent laboratory analysis, on the numerical concentration of particles and their size and shape.

- Surface concentration

The ELPI, which was described above, is capable of real-time detection, based on size, of the surface concentration. The SMPS and the FMPS can establish the size which can be related to the surface of particles, under certain conditions. Electron microscopy also has the possibility of providing certain information on the surface of the particles in relation to their size. The diffusion battery allows real-time measurement of the surface of the aerosol particles when coupled to an appropriate preseparator. The epiphaniometer, developed specifically for the measurement of nanoparticle, is another instrument capable of measuring the total surface of particles between 10 and 1000 nm.

- NP characterization based on size

The SMPS, which is currently the instrument most widely used for the determination of NP, was covered in the section discussing the mass concentration of NP. The FMPS, a new-generation instrument, should be able to obtain the same type of information as the SMPS, but with a quicker response time. The SMPS is able to determine NP granulometric distributions from 3 to 1000 nm (ISO, 2007a; TSI, 2009), with a rapid response time, and presents the advantage that the mobility diameter is almost equivalent to the projected particulate surface diameter. It thus can estimate not only the granulometric distribution but the particulate surface area. On the other hand, it is difficult to use in the work environment because of the size of the instrument, its cost, the fact that several instruments may be necessary to cover the entire range of granulometries to be evaluated, and finally its complex operation. Its radioactive source could require a trip permit in certain situations.

The diffusion batteries can determine the continuous granulometric distribution of an aerosol based on the measurement of the NP diffusion coefficient, which can be converted into the NP diameter.

Used serially with a condensation nuclei counter, these batteries make it possible to determine a granulometric distribution by number of particles.

Cascade impactors, also described above, have the possibility of determining the granulometric distribution of an NP aerosol by inertial deposition, with different aerodynamic cut-off diameters. Several models exist that allow personal sampling, but in granulometric domains greater than 200 nm. However, cascade samplers are available for NP, with a minimum cut-off diameter of 10 nm, but only for fixed station collection. Given that the cascade impactors measure the mass granulometric distribution of NP, normally unquantified hypotheses regarding their shape and density must be elaborated to estimate the granulometric distribution by number of particles or surface area.

The electrical low pressure impactor (ELPI) allows inertial collection coupled to electrical detection in almost real time for particles bigger than 7 nm. Thus, each stage of the impactor, linked to a narrow range of aerodynamic diameters, is attached to an electrometer to measure the accumulated charge directly associated with the active surface. If the efficiency of aerosol charging is known, the ELPI data can establish the granulometric distribution by number of particles and by mass. Since the ELPI allows accumulation of particles on the impactor's different levels, they could be analyzed subsequently by complementary techniques, such as microscopy, to confirm their size, number or shape, or chemical analyses capable of searching for an atom, such as a metal specific to the NP.

- Other types of NP characterization

It may prove important to determine the physical and chemical properties of NP, which can be linked to human health effects. Parameters such as size, shape, surface, composition, crystallinity, solubility and biopersistence provide basic information on exposure and toxicological assessment of new nanomaterials. Surface coating and electrical charge will have a significant impact on the aggregation level, which in turn will influence physical behaviour, the pulmonary deposition site and ultimately the biological response. However, the determination of these parameters and the study of the surface properties exceed the objectives of this document. They will not be discussed here.

- The need for developing new measuring instruments

Several research groups and companies are actively working on the development of portable instruments for directly measuring the surface of NP. For example, TSI has been offering for a short time now the AEROTRAK™ 9000 *Nanoparticle Aerosol Monitor*, which is a diffusion battery coupled to an electrometer. This portable instrument, powered by rechargeable batteries, allows the direct reading of the surface of particles from 10 to 1000 nm in size that could deposit in the tracheobronchial and alveolar regions of the lungs. Fissan *et al.* (2007) developed a method for determining in real time the total specific surface of NP from 10 to 100 nm by modifying an electrical detector for aerosols with an ion trap already used for routine examinations in workplaces. The current obtained is proportional to the surface of the particles studied. The instrument provides a direct surface measurement for NP larger than 10 nm with a range of application from 0 to 10,000 $\mu m^2/cm^3$ (Shin *et al.*, 2007). Other instruments are also being developed, evaluated, or are already available commercially. These include an instrument using the

ionization technique coupled with an optical approach (Litton *et al.*, 2004; Edwards *et al.*, 2008), the Nano-ID developed by Particle Measurement Systems (2009), and the NanoCheck (Spielvogel *et al.*, 2009).

The question about the necessity of developing new instruments adapted to the specific characteristics of NP to make up for the shortcomings of existing measuring instruments in occupational hygiene is actually the subject of consensus in the scientific community. It is similar to the question, as a corollary, about the development of multiple NP standards that can be used for instrument calibration (Stefaniak *et al.*, 2009).

8. NANOPARTICLE EXPOSURE ASSESSMENT

Occupational hygienists have different reasons for sampling NP:

- personal exposure assessment in terms of compliance with the standards in force or with a specific action threshold for implementation of control measures;
- personal exposure assessment eventually allowing exposure to be linked to health effects;
- identification of the main emission sources to establish an emission control plan;
- assessment of the effectiveness of the control measures put in place.

The assessment strategies and the choice of collection and analysis techniques then must be adapted to the specific objectives of the intervention. Assessment of occupational NP exposure of the breathing zone should allow measurement of the aerosol NP parameters associated with the inhalation health risks and thus favour characterization of the particles dispersed in the aerosol. Whenever possible, this assumes the use of personal collection methods from portable instruments installed in the worker's breathing zone (Agence nationale de recherche, 2005).

Consequently, it is appropriate to think about the possible approaches to achieving a better characterization of NP in a context in which several of the instruments available reveal significant limitations, making it difficult to measure some parameters directly. Among the main limits on occupational exposure assessment, we should mention the unsuitability of most instruments to measurement in the breathing zone and their inability to accumulate data for the entire workshift. The intrinsic properties of NP, such as the tendency to agglomeration, may also lead to substantial spatiotemporal alterations of the aerosols inhaled by the worker in terms of the mass, number and granulometric distribution of the particles, the lung deposit site or even their toxicity.

It has been clearly shown that measuring mass concentration alone is not enough to characterize NP (see Chapter 6). As a minimum, it is also essential to assess the particle concentration in number and surface area and the granulometric distribution. It would be no less prudent to establish the current aerosol mass exposure by granulometric fraction to have maximum information allowing exposure characterization.

The ISO standard (2007) mentions that no perfectly appropriate granulometric selector, with a 100 nm cut-off diameter is currently available on the market. Worker exposure measurements are usually taken in the breathing zone. However, they can be deduced very prudently from samples collected at fixed sites. Indeed, notable variations of numerical and granulometric concentration have been reported in the literature, even for short distances (variations in time and according to the distance from the source) (Brouwer *et al.*, 2004). These authors also conclude that personal exposure concentrations are normally higher than fixed site environmental concentrations.

As already mentioned, two major factors contribute to the changes in properties observed for NP compared to more voluminous materials: a much greater relative surface per unit of mass and a predominance of quantum effects. The first factor is responsible for reactivity changes, while the second, observed for particles of a few tens of nm, is responsible for changes in optical, electrical, electronic and magnetic properties. Indeed, 30 nm particles have 5% of their atoms on the surface, compared to 20 % for 10 nm particles and nearly 50% for 3 nm particles. The hygienist thus will have the challenge of producing an adequate assessment of occupational exposure.

Airborne sampling strategy

Fixed site sampling does not account for the workers' different activities, their location in relation to the emission sources, their work methods and any other factor which can influence their exposure. The choice of sampling site (or sites) is of primary importance and must account for factors including emission sources, occupational activities, air currents and other particles present in the workplaces capable of influencing measurements. The instruments currently available are normally not NP-specific and consequently produce results for all the aerosols present. Nonetheless, while accounting for the limits of this approach, a good assessment strategy can allow an occupational exposure estimate. The combined use of fixed site instruments and short-term individual sampling, as part of an adapted sampling strategy, could provide very useful information on NP exposure.

To obtain a good idea of airborne NP concentrations, several organizations and authors (Brouwer *et coll.*, 2004; NIOSH, 2006, 2008a, 2009b; ISO 2007, 2008a, 2008b; AFSSET, 2006; Ostiguy *et coll.*, 2006b, 2009; Harford *et coll.*, 2007; OCDE, 2009; Kuhlbusch *et coll.*, 2004, 2006, 2008, 2009; Chow et Watson, 2007; Maynard et Aitken, 2007; HSE, 2006; CSA, 2010.) propose a sampling strategy that simultaneously incorporates several methods and calls on different ways of measuring NP. To find out workers' exposure, they recommend that the main NP characteristics be documented.

Quantitative exposure assessment currently poses an additional challenge, because every environment already contains an often complex mixture of UFP present in the indoor and outdoor air of industrial establishments. Both in rural and urban settings, environmental air can contain millions of particles of nanometric dimensions per litre of air. Through this mixture of dust particles of different granulometries and various compositions, it is essential to determine the content and characterize the manufactured NP, in a context where the natural background noise can substantially contribute to the instrument reading.

Another important element in exposure assessment is to determine the desired granulometric fraction. Indeed, the behaviour and deagglomeration capacity of agglomerates is a determining phenomenon in pulmonary toxicity, but one that currently is relatively undocumented. In the case of complete deagglomeration, it is logical to believe that the biological impact would be identical between the agglomerate and the individual particles, for the same total mass and the same lung deposit site. Consequently, even though individual NP, by definition, are smaller than 100 nm, agglomerates may be substantially more voluminous and their exposure would need to be documented.

The OECD (2009) recommends starting the evaluation by identifying potential NP emission sources from a review of the type of process applied and the different operations, the materials involved as well as the tasks and work methods. The information available (MSDS, literature, etc.) is analyzed in order to better understand the characteristics of the NP produced or used: chemical composition, size, shape, solubility, reactivity, toxicity, explosiveness, flammability, etc.

Once the potential emission sources have been identified from a review of the process and the available information, the hygienist:

- conducts an observational walkthrough survey of the production area and processes to locate potential sources of emissions;
- determines the frequency and duration of each operation and the personnel involved;
- documents the type of equipment used for handling NP and identifies potential leakage sources likely to promote the emission of NP;
- documents the implemented means of emission control (insulated room, closed circuit, enclosure, ventilation at source and general ventilation, etc.), data on performances and their maintenance, and potential leakage sources;
- documents the process zones where containment is deliberately breached: opening of access door for preventive maintenance, repair, cleaning, etc.

The first step of an assessment strategy is to identify the potential outdoor UFP sources and all the other sources of UFP in the workplace contributing to background aerosol concentration, and susceptible to mask the presence of engineered NP. In the second place, studying the ventilation (air change rate), movements and air currents and the presence of aerosols will provide indispensable information on the transport of environmental particles (natural background), and of engineered NP. This is why the use of smoke tubes is not recommended, given that the release of small particles into the environment could distort the collection results (Brouwer *et al.,* 2004). Thirdly, before measuring NP released by the processes, it is necessary to estimate the ambient UFP concentration to know the background aerosol concentration in the work environment. Finally, study of workers' movements should help find the best location to place the measuring instruments. Then it only becomes relevant to quantify the emission sources and types of particles released or transported. Most of the resuspended dusts should consist of relatively large particles which, in return, could play a role in future airborne NP. The fixed site measuring instruments must be placed strategically for the most precise idea possible of workers' exposure.

NIOSH (2008a), the OCDE (2009) and the CSA (2010) suggest the initial determination of the background at different locations and workstations close to the equipments **before** starting the processes producing or using NP. They suggest determining the number of particles of small size by means of a CNC (condensation nucleus counter), and of larger size with an OPC (optical particle counter). If there is a large number of measured particles (the values are relative and a function of the process and site), it would then be appropriate to check for the presence of a source of particle generation in the environment: natural gas consumption units, vacuum pumps, gasoline/diesel/propane lift trucks, combustion sources or heat or atmospheric pollutant generating activities such as welding or heat sealing. The recirculation of unfiltered air could also become a source of UFP, just like the resuspension of deposited particles following activities involving the movement of equipment and personnel. Measurement of the aerosol level will be reevaluated **after** operations involving NP. The interpretation of measurements of the NP level during operations will take into account these control values before and after the operations. In the situation where the background levels are stable before and after the NP are handled, and the NP levels measured during the operations are high compared to this background level, then the concentration of NP generated by the application of different processes can be determined from the difference.

During operations involving NP, the concentration of the airborne particles and their dimensions are measured simultaneously with a CNC and an OPC close to the identified or potential emission sources (e.g., opening of reactor, packaging, handling of powders). Parallel sampling on filters for

subsequent electron microscopy analysis is also recommended in order to confirm the presence of NP and to determine their shape. The concentrations are determined **before, during** and **after** each task or operation in order to identify the factors that could affect the concentrations of airborne particles. This information will then be used to determine the situations where additional complementary samples will be required in order to properly characterize the exposure.

All this preliminary information is necessary for defining a detailed strategy for evaluating worker exposure. In the current context, in which scientific knowledge cannot establish clearly which parameter is the best able to correlate NP aerosols to their toxicity, Warheit (2009), SCENIHR (2009) the OCDE (2009), and ISO (2008a, 2008b) recommend accumulating the maximum information on exposure concentrations. Incidentally, the instruments should be reasonably priced, transportable and robust (Maynard and Kuempel, 2005).

Table 6, inspired, among other sources, by technical reports from ISO (2007, 2008a, 2008b), AFSSET (2008), and Harford *et al.*, (2007) lists some examples of instruments and techniques allowing characterization of NP aerosols. For more detailed information, Chow and Watson (2007) produced an exhaustive review of all the ultrafine particle measuring methods available and applicable to the NT field.

Table 6 : Examples of Instruments and Techniques Allowing Characterization of NP Aerosols

Parameter	Instruments	Remarks
Direct mass measurement	Cascade impactors	Berner cascade impactors or MOUDI or Nano-MOUDI Micro-Orifice Uniform Deposit Impactors allow gravimetric analysis of stages finer than 100 nm during individual assessment.
	TEOM	The Tapered Oscillating Element Microbalance (TEOM) preceded by a granulometric selector can be used to determine the mass concentration of nanoaerosols. This method is sensitive and allows real-time measurement.
	Filters	Filters, equipped with appropriate preselectors as needed, can be used to sample NP to determine the mass or to perform subsequent laboratory analyses.
Mass estimate	ELPI	Real-time detection by size and active surface concentration gives a granulometric distribution of aerosol particles. If the charge and density of the particles are known or assumed, the data then can be interpreted in terms of mass concentration. The samples from each stage then can be analyzed in the laboratory. Lower limit: 7 nm.
	SMPS / FMPS	Detection of the numerical concentration based on size and real time gives a granulometric distribution of the aerosol. Knowledge or estimation of the shape and density of the particles then makes it possible to estimate the mass concentration.
	MOUDI/nanoMOUDI	The Micro-Orifice Uniform Deposit Impactor (MOUDI) can determine the aerodynamic diameter by cascade impaction.
Direct number measurement	CNC	Condensation nucleus counters (CNC) allow real-time measurement of the numerical concentration of particles within the particle diameter detection limits. Without a granulometric selector, this instrument is not specific to the nanometric field. The TSI P-Trak offers preselection with an upper limit of 1000 nm and a lower limit of 20 nm. Using the TSI 3007, the lower limit of detection drops to 10 nm.

Parameter	Instruments	Remarks
	Optical counters	Optical counters give a real time measurement of particles with diameters from 300 nm to 10,000 nm. These instruments, which do not detect individual NP, can nevertheless be very useful for determining agglomerates of NP and measuring the background of the work environment.
	SMPS and FMPS	The Scanning Mobility Particle Sizer (SMPS) Spectrometer and the Fast Mobility Particle Sizer (FMPS) Spectrometer allow real-time detection based on the electrical mobility diameter (related to size) and the numerical concentration.
	Electron microscopy	Analysis by electron microscopy can provide information on the shape, granulometric distribution and the numerical concentration of the particles constituting the aerosol.
Estimating the number of particles by calculation	ELPI and MOUDI/nanoMOUDI	Real-time detection by size or active surface concentration gives a granulometric distribution of aerosol particles. If the charge and density of the particles are known or assumed, the data then can be interpreted in terms of number of particles. The samples from each stage then can be analyzed in the laboratory.
Direct specific surface measurement	Diffusion battery	Commercially available diffusion batteries allow real-time measurement of the aerosol's active surface and have a response in relation to the active surface of particles smaller than 100 nm. These instruments are NP-specific if they are used with an appropriate pre-separator. The TSI Aerotrak 9000 is an example of an instrument that allows real-time measurement of the surface and the concentration.
	ELPI and MOUDI/nanoMOUDI	The ELPI allows real-time detection of the aerodynamic diameter according to the size and the active surface concentration. The samples from each stage then can be analyzed in the laboratory.
	Electron microscopy	Analysis by electron microscopy can provide information on the surface of the particles in relation to their size. Transmission electron microscopy provides direct information on the projected surface of the particles analyzed, which can be linked to the geometric surface for certain shapes of particles.
Specific surface by calculation	SMPS / FMPS	The SMPS and the SMPS allow real-time detection based on the electrical mobility diameter (related to size) and the numerical concentration. Under certain conditions, the data can be interpreted in terms of specific surface.
	SMPS and ELPI used in parallel	The differences in measurement of aerodynamic diameters and mobility can be used to deduce the fractal dimension of the particle, thus allowing subsequent particle surface area estimating.

HSE (2006) emphasizes the importance of evaluating the performance of instruments in the laboratory before their use in the work environments, because abnormal responses and discrepancies were observed, particularly in mobility analyzers with ultrafine CNT and agglomerates or with NP < 10 nm.

In its NanoAlerts series (2006, 2007a, 2007b), the HSE tracks new analytical developments. Table 7 summarizes a few microscopic techniques that can be used (HSE 2006).

Other laboratory instruments can also contribute to NP characterization, including flame or graphite furnace atomic absorption spectroscopy and inductively coupled plasma mass spectrometry (ICP-MS).

Table 7 : Types of Electron Microscopes which Can Contribute to NP Characterization

Parameter	Instruments	Remarks
Dimension, shape, surface, chemical composition	Transmission electron microscopy (TEM)	The shape and granulometric distribution can be determined by the fractal dimension of the agglomerates. The individual NP composition can be documented by coupling the TEM to energy-dispersive X-ray spectroscopy (EDX) or electron energy loss spectroscopy (EELS). High resolution microscopes can analyze particles smaller than 1 nm.
	Scanning electron microscopy (SEM)	The shape and granulometric distribution can be determined by the fractal dimension of the agglomerates. The individual NP composition and the average composition (mapping) can be documented by coupling the SEM to energy-dispersive X-ray spectroscopy (EDX) or electron energy loss spectroscopy (EELS). High resolution microscopes can analyze particles smaller than 1 nm.
Dimension, shape, surface, chemical composition	Field emission gun-scanning electron microscopy (FEG-SEM)	The shape and granulometric distribution can be determined. The fractal dimension of the agglomerates can be determined with high-resolution FEG-SEM. The individual NP composition and the average composition (mapping) can be documented by coupling the FEG-SEM to energy-dispersive X-ray spectroscopy (EDX). High resolution microscopes have a resolution of around 1-2 nm.

Cutaneous sampling strategy

Bibliographical research did not allow identification of articles dealing with the problem of assessing cutaneous NP exposure. The established cutaneous sampling methods must therefore be adapted to other chemical substances (Hoang, 1992; Brouwer *et al.*, 2000; Viau *et al.*, 2004; CEN, 2005; Ignacio *et al.*, 2006).

Cutaneous exposure assessment can be performed by measuring the quantity of NP in contact with the skin for a certain time period. The existing methods are based either on recovery of dusts accumulated on the skin, or by interception of this dust during contact. However, the first method may raise uncertainty related to the difficulty of quantitative dust recovery and necessitates an evaluation of the contact time. The precision of the interception method is likely to be affected by the adherence characteristics of the collection material, which may not mimic the interactions with the skin exactly.

Recovery of dusts deposited on the skin

Three main experimental approaches allow recovery of NP deposited on the skin:

- Several solvents can be used to wash the skin and the solutions obtained can be analyzed subsequently;
- Solvent-impregnated materials can also be used to wash the skin and the recovered dusts are analyzed subsequently;
- The use of adhesive paper applied repeatedly to the skin ensures recovery from the surface and the superficial layers of the skin (Tsuji *et al.*, 2006).

Regardless of the recovery method, the collected samples are then characterized by using a whole battery of laboratory tests, particularly those discussed previously.

Dust interception before cutaneous deposit

Three main experimental approaches allow interception and capture of NP before they are deposited on the skin:

- Various materials (cotton, polyester, polyurethane foam, etc.) can be applied to different places on the body to collect NP dusts. This approach remains qualitative. In general, capture efficiency is considerably lower by recovery than by interception. These approaches reflect the quantity of dusts accumulated better than the exposure level;
- Absorbent gloves can also allow capture of NP likely to come into contact with the hands. Maynard (2004) used cotton gloves, which were slipped over the worker's gloves, to assess the CNT concentrations to which the skin is exposed.
- Dosimetry of the entire body can be considered by using coveralls; the main deficiency is the difficulty of recovering the NP trapped in all this fabric.

8.1 Risk of Occupational Exposure during Nanoparticle Synthesis by Conventional Processes

Any evaluation of exposure to particles should follow a rigorous procedure by applying the guiding principles described in chapters 8, 20 and 21 of the Occupational Hygiene manual edited by Roberge *et al.* (2004). To study the risks associated with NP exposure, several factors which can influence exposure must be considered, including the risks related to the raw materials used, the intermediate products formed during the processes and the end products. All the production steps, from the arrival and storage of the starting materials to the shipping of the final product, must be examined. The state of the material (fine powder, granular, in suspension, in a liquid, etc.), the processes used (in closed reactors under vacuum, in open tanks, production in gas or liquid phase, etc.), the degree of confinement at each step in the process including collection, handling and packaging of NP the total quantity of material handled, the possibility of aerosolization and the ability of particles to become airborne or land on work surfaces, ventilation efficiency, workers' exposure time, work and storage methods, and waste management are a few of the parameters to be retained.

There is an exposure risk for the vast majority of NP production processes, including synthesis, recovery and handling of synthesized materials (ICON 2008). The risk level can vary enormously depending on the specificities of the work environment. During synthesis, exposure can be caused by a leak or by frequent starting and stopping of the process (Biswas and Wu, 2005). On the other hand, the nature and level of exposure are likely to differ greatly depending on the process used and its stage. The means of prevention implemented must therefore take these variables into account. Referring to the major process classes described above, Table 8 summarizes the main exposure risks during NP synthesis (Aitken *et al.*, 2004; ICON 2008).

Table 8 : Risks of Exposure to Nanoparticles during their Synthesis

Synthesis process	Inhalation risks caused by or during	Cutaneous absorption or ingestion risks caused by or during
Gas phase condensation	Reactor leak Product recovery Post-production treatment Bagging, unbagging Shipping Maintenance / cleaning of production and ventilation sites and equipment Accidental spill	Resuspension Sedimentation Product handling Shipping, maintenance / cleaning of production and ventilation sites and equipment Accidental spill
Vapour deposition	Reactor leak Product recovery Post-production treatment Bagging, unbagging Shipping Maintenance / cleaning of production and ventilation sites and equipment Accidental spill	Resuspension Sedimentation Product handling Shipping, maintenance / cleaning of production and ventilation sites and equipment Accidental spill
Colloid formation and chemical precipitation processes (low risks)	Product drying Spill / drying Agitation / transfilling Maintenance / cleaning of production sites and equipment	Spill Solution handling Shipping, maintenance / cleaning of production and ventilation sites and equipment Accidental spill
Mechanical attrition processes	Reactor leak Product recovery Post-production treatment Product recovery Bagging, unbagging Shipping Maintenance / cleaning of production and ventilation sites and equipment Accidental spill	Spill Product handling Shipping, maintenance / cleaning of production and ventilation sites and equipment Accidental spill

In gas phase processes, NP are found in suspension in a gas in reactors. During a leak, NP can escape, especially if the reactor is operated under positive pressure. The nature of the aerosol NP then would depend on the location of the leak. At the beginning of the process, primary NP especially could be released into the air. Subsequently, these NP would appear increasingly in aggregated or agglomerated form. At the end of the process, agglomerated NP could be emitted in the work environment (Aitken *et al.,* 2004).

There is little likelihood that vapour phase deposition processes release NP during the synthesis phase because they form directly on a substrate by vapour deposition in a controlled thickness

film. In some processes, the product is recovered from the substrate by an automated or manual mechanical process. It is then possible that agglomerates will leak into the ambient air.

Gas phase and vapour phase deposition processes may cause exposure, especially if the workstation is not isolated (confined) and if NP can disperse and thus expose workers in other sectors.

In some chemical processes, colloids are formed in solution so that inhalation exposure during the synthesis process is less probable. Inhalation exposure of a worker affected by a wet process (colloid formation) would occur mainly during agitation and transfilling (NIOSH, 2007, 2009b). An inhalation risk then would exist via aerosol droplets containing NP, although we had little information on the subject at that time (Maynard and Kuempel, 2005). In some processes in which the product is recovered by vaporization in an evaporation chamber, there is a risk of inhalation exposure to agglomerated NP, especially in case of a leak.

Several processes can be used to produce NP from larger sized particles. Some of them crush large particles until they reach nanometric dimensions. These operations, particularly if they are done dry, can generate significant quantities of inhalable particles. Just the handling of powders can release particles into the air. They must therefore be handled and stored properly.

In general, recovery, post-recovery treatments, as well as the packaging of a dry finished product and equipment maintenance represent some potential sources of exposure. Depending on the efficiency of the existing recovery system, individual NP, aggregates or agglomerates can escape and be found in the ambient air, and then be carried by the ventilation system. Nonetheless, the inhalation exposure risk will depend on the process used, the product's characteristics and its airborne resuspension potential.

Regardless of the approach to NP production, there is always a risk of exposure via the cutaneous route or ingestion when a worker touches contaminated surfaces following spills or atmospheric emissions, or when he handles the product during packaging, shipping or at the time of a spill. Equipment maintenance work and general maintenance of the workplace are other occupational exposure situations where, both pulmonary and cutaneous exposure can occur (NIOSH, 2007, 2009b). Colvin (2003b) adds that many NP are prepared and handled in water, which can cause cutaneous or digestive absorption. Poor personal hygiene will favour NP ingestion (Aitken *et al.*, 2004).

In conclusion, rigorous management and risk reduction at the source are required to limit the occupational exposure risk. Workers, like researchers and their students, must be very prudent when handling NP, whether in production, handling, conditioning, transferring, packaging, storage, receiving or use of these products, scrap disposal or maintenance and cleaning of sites and equipment. Exposure to dusts containing NP is also possible during operations on nanomaterials: cutting, machining, sanding, etc. Several work situations could require the wearing of personal protective equipment (see Chapter 9). A best practices guide was developed to facilitate the assumption of responsibility for identification and management of different health and safety risks for workers and researchers (Ostiguy *et al.*, 2009).

8.2 Measurement of Occupational Exposure during Synthesis of Nanoparticles by Conventional Processes

Currently there are a limited number of published studies for estimating or determining the level of occupational exposure to NP. In this sense, HSE, in collaboration with the Health and Safety Laboratory (HSL), produced an excellent literature review at the end of 2006 (HSE, 2006). At that time, these authors had identified fewer than 10 studies published since 2000 on NP occupational exposure or dispersion: these studies dealt with exposure to carbon black, inks, titanium dioxide, silica fumes and CNT (HSE, 2006). Updates were produced twice in 2007 (HSE 2007a, 2007b). Among these studies, HSE (2006) reports a study by Moehlmann (2005) in different settings: offices, silica fusion, plasma torch cutting, metal grinding, pastry shops and airports. In an assessment program begun in 1998, the Institute for Occupational Safety and Health of the German Berufsgenossenschaften (BGIA) and the Institute for Hazardous Materials Research (IGF) obtained UFP levels in concentration fields from 108 to a few thousand particles measuring from 10 to 500 nm/cm^3. These outcomes included agglomerates. In another study of fine carbon black, fine nickel powder, precious metal black with high specific surface, titanium dioxide, metal fumes and metal oxides, zinc and zinc oxide coming from metallurgical and refining processes, welding fumes and steel smelters, Wake (2006) reports that the concentrations found outdoors were equal to or greater than those encountered near workers operating the processes, including those in the packing section. During some light metal or carbon pellets packing operations, the concentrations could increase substantially. Remember that the direct reading instruments used in the work environments currently cannot discriminate between NP and UFP.

In 2006, Kuhlbusch *et al*. reported carbon black UFP measurements in three plants. One plant did not show any excess number of particles compared to outdoors, while a second plant had peaks in the reactor and pellets formation sections, although these peaks could be linked to outdoor vehicular traffic. Only the third plant showed high concentrations of ultrafine particles, also in the reactor and carbon pellets formation section. The reactor was in closed circuit operation and the authors hypothesized that the dusts measured could be linked to the oil and grease fumes emitted during maintenance operations. In the pellets section, the authors conclude that the high number of UFP is linked to a leak in the production line.

Maynard *et al.* (2004), to our knowledge, are the first to have studied certain characteristics of exposure to single-walled CNT subjected to mechanical agitation, first in the laboratory and then in the work environment, to determine the pulmonary and cutaneous exposure risk factors. Two synthesis techniques were studied, which lead to production of single-walled CNT, metal catalyst particles and other forms of elementary carbon, all of nanometric dimensions. Under the specific conditions of their study, Maynard *et al.* (2004) reported air concentrations of CNT between 0.7 and 53 μg/m^3. Examination of the samples showed that several of the particles were compact rather than having a low-density structure. Also noted were clusters of non-inhalable materials. Their estimation of the total CNT found on the gloves that were used for the various manipulations varied from 217 to 6,020 μg, with most of the material being found on the parts of the glove in direct contact with the surfaces.

By applying the same experimental conditions in the laboratory to alumina fumes, the airborne concentration generated exceeds that of CNT by about two orders of magnitude (Maynard *et al.*, 2004). In a more recent study, Han *et al.* (2008) measured MWCNT concentrations as high as 0.4 mg/m^3 containing 194 MWCNT/cc in a laboratory making CNT. The installation of control measures made it possible to reduce the concentration to undetectable gravimetric levels and to 0.05 MWCNT/cc. Fujitani *et al.* (2008) assessed fullerene exposure in a production plant. They measured few modifications of particle concentrations of nanometric dimensions before and during the process. However, during weighing and packaging, they measured a higher concentration of particles bigger than 1,000 nm. The use of a vacuum cleaner increased the concentration of particles <50 nm but not the concentration of particles > 1 000 nm. An examination by electron microscopy showed that the biggest particles consisted of fullerene aggregates. They could not reach a conclusion on the origin of particles < 50 nm.

These studies provide preliminary indications of the propensity of CNT to form nanometric aerosols during the process. It is prudent to reduce workers' pulmonary and cutaneous exposure risk, provided that these fine particles normally contain metals used as catalysts, particularly nickel, which is recognized as carcinogenic. Carbon black aerosol emission measurements during bagging operations showed mass concentrations of PM10 up to 20 times higher than the ambient concentrations, with most particles having diameters bigger than 400 nm, while particles < 100 nm were mainly attributed to sources other than carbon black (Kuhlbusch *et al*, 2004). Gray and Muranko (2006) studied the behaviour of carbon black and amorphous silica agglomerates. They concluded that a severe mechanical process caused a midscale breakdown of the biggest aggregates and released very few elementary particles. Hsu and Chein (2007) studied the emission of TiO_2 nanopowder, which had been used as a coating material on different substrates. They proceeded with accelerated aging tests (ultraviolet radiation, fan, human contact simulated by a rubber knife). The highest emissions were measured on tiles coated with titanium dioxide (22,000 particles of 55 nm/cm^3). The emission rate for the tile coating increased again after two hours of experimentation, while it had decreased substantially after 60 minutes on polymer film and after 90 minutes on wood.

Demou *et al.* (2008) used a gas phase process to estimate the airborne NP concentration (unspecified nature) in a pilot production unit. The study confirmed that the highest exposures were in the production unit and that the NP concentration pattern was directly correlated to the process phases. The maximum concentration was equivalent to about four times the UFP concentration measured when the process was not in operation. Vacuum cleaning also represented a situation in which the airborne dust level increased. However, handling of particles and the process during postproduction operations did not lead to a significant increase in the particle level.

8.3 Occupational Exposure during use of Nanoparticles

The use of NP in aerosol-generating processes can increase the risk of inhalation or skin absorption. The spraying of disinfectants, air fresheners, paints, dyes and products to impregnate fabrics or porous materials are some occupational exposure situations where significant amounts of NP can be emitted into the air (Hett, 2004). The steps in handling powders, mainly regarding the reception and application of NP, are other potential exposure situations, but the studies allowing these exposures to be characterized and quantified are still very limited.

Powders have a natural tendency to become suspended in the air, particularly if they are finely divided. Some nanopowders are specially treated to prevent the formation of agglomerates. The potential for air dissemination and for long-term suspension is then greatly increased. Highly reactive particles or ones with a long biopersistence period can be particularly hazardous for an exposed worker. Schneider *et al.,* (2007) report that handling nanomaterials generates dust particles, which vary in quantity and granulometric distribution according to their handling scenarios and the properties of the materials. Several dust generation protocols have been developed, using different approaches to simulate actual nanomaterial handling conditions. In particular, the dust generation level is a key determinant in explosion risk assessment. At this time, however, there does not seem to be a consensus on the best test to use. The adoption of a test by a laboratory nonetheless would allow comparison of the dust generation rate for different NP under equivalent experimental conditions. Thus, Schneider *et al.,* (2007) have developed a new test and report the results of a few laboratory tests. Under identical experimental conditions, 18.6 nm TiO_2 particles generated about 300 times more dust than 170 nm particles. A much higher exposure risk can thus be forecast for TiO_2 NP compared to larger NP. Consequently, it is necessary to institute stricter safety procedures if larger particles are replaced with smaller particles.

The use of NP, like their production, necessitates rigorous management and reduction of the factors to limit the occupational exposure risk. Workers must therefore be very careful in handling NP, whether it involves their storage, use, or the elimination of waste or even the maintenance of equipment and the premises. Aitken (2004) had reported high exposure levels over short periods during certain maintenance operations. Indeed, suspension or resuspension of NP during handling and maintenance can contribute to workers' respiratory and cutaneous exposure risk. Equipment and work activities can also trigger resuspension of airborne particles, just like vibrating pipes or movement of workers or vehicles. Wearing personal protective equipment thus should be required in several work situations, particularly for maintenance or as long as the effectiveness of that at-source elimination and ventilation measures implemented is not proved.

Recently, Bello *et al.* (2009) assessed the exposure to particles of nanometric dimensions and fibres during dry and wet machining of composites containing CNT. Wet machining did not significantly increase the exposure level compared to the environmental concentrations already present, and exhaustive electron microscopy research did not reveal the presence of CNT. However, dry machining led to concentrations of 2 to 4 fibres per cubic centimetre, depending on the type of composite used.

Peters *et al.* (2009) used two approaches to differentiate the concentration of NP in the workplace from that of the other airborne particles in a company producing metal oxides of lithium and titanium. By specifically analyzing the metals involved by ICP, the researchers determined that the concentration of respirable dusts was low (< 50 ng/m^3) and contained less than 10% titanium, and therefore mainly consisted of atmospheric particles at all workstations except where the NP were extensively handled. The mass concentrations could then go as high as 118 ng/m^3 and contain up to 39% lithium and titanium. Electron microscopy analysis determined that the sampled particles consisted of agglomerates of NP larger than 200 nm made up of elementary particles from 10 to 80 nm (Peters *et al.,* 2009). These researchers also used direct-reading instruments to determine the concentrations of particles < 300 nm. They concluded that the measured concentrations did not show an apparent correlation with the work activities, but that the concentration peaks coincided

with the loading of supply bins or the replacement of nanomaterial filter bags. They concluded that the concentrations of airborne NP are dominated by point sources.

Geraci *et al.* (2009) used direct-reading instruments (condensation nucleus counters and optical counters) to evaluate the exposure of workers in approximately a dozen American companies that produce and use NP, and also collected samples on filters for subsequent laboratory analysis. These studies made it possible to document the occupational exposure level and to demonstrate that the usual means of controlling exposure can be very effective. The results of several studies to determine occupational exposure levels were recently presented at the 4th International Conference on Nanotechnology Occupational and Environmental Health held in Helsinki from August 26-29, 2009. The levels of NP exposure are extremely variable, depending on the processes used and the phase of the NP: solid or in solution (Riediker *et al.*, 2009; Brouwer *et al.*, 2009; Mark *et al.*, 2009; Berges *et al.*, 2009; Ichihara *et al.*, 2009; Tsai *et al.*, 2009; Chou *et al.*, 2009; Huang *et al.*, 2009a, 2009b; Shinohara *et al.*, 2009; Koivisto *et al.*, 2009; Jarvela *et al.*, 2009; Kim *et al.*, 2009; Evans *et al.*, 2009; Birch *et al.*, 2009; Mohlmann *et al.*, 2009; Ono-Ogasawara *et al.*, 2009). Also, the European observatory (Kaluza *et al.*, 2008) summarized the knowledge on the levels of workplace exposure by placing particular emphasis on the NP of silica, iron and silver, as well as on single-walled CNT. For unpurified single-walled CNT, the authors (Kaluza *et al.*, 2008) reported that large aggregates visible to the naked eye are generated in the laboratory but that they contain very few small particles. Laser ablation synthesis processes generate few particles smaller than 100 nm, while the process using carbon monoxide at high pressure generates particles smaller than 100 nm that have not been characterized. It is therefore impossible to determine whether they are CNT, particles of catalyst, or non-fibrous carbonaceous particles. Finally, we should mention that Kaluza *et al.* (2008) reported that the measured generation of single-walled CNT aerosols was two orders of magnitude smaller than that for alumina fumes under the same experimental conditions. These authors also reported the evaluation of the air contamination level in four companies that handled CNT. Kaluza *et al.* (2008) concluded that there was no clear evidence of an increase in mass concentration of aerosols during the handling of unrefined CNT.

9. EXPOSURE PREVENTION AND CONTROL

A best practices guide to working safely with NP was published recently by our team (Ostiguy *et al.*, 2009). This guide provides a detailed description of the steps for assessing and managing different kinds of risks and proposes a structured approach to characterize and control them, while considering uncertainties related to toxic, fire and explosion risks and the workers' exposure level. The guide proposes a development and implementation approach for a workplace prevention program. This chapter therefore will be limited to recapitulating some of the points contained in the guide and to summarize the current knowledge on the efficiency of exposure control measures. For more information, the reader is invited to read the best practices guide (Ostiguy *et al.*, 2009) and the Work Safe Australia document (2009a) on the efficiency of exposure control measures.

Risk assessment is an essential preliminary step to determine what control level must be implemented to limit emissions of NP and prevent a toxic substance from affecting certain target organs in workers. Thus, the control measures must be proportional to the documented, estimated or potential risk and the uncertainties related to these risks. However, in the absence of adequate knowledge of the toxicity and behaviour of airborne nanoparticles, and the absence of specific standards, strict control measures should be put in place to minimize, as much as possible, the risk to workers of pulmonary and cutaneous absorption. In the meantime, the knowledge acquired in control of UFP can serve expediently as a guide to the NP field.

With nanoparticles that can become airborne and inhalable, there may be a potential for occupational exposure, whether in production, use, equipment maintenance, recycling or waste disposal. This occupational exposure is currently considered to be the leading source of population exposure (Royal Society, 2005). Among the major factors contributing to NP exposure in the work environment, we should mention the quantity of product handled, the duration of exposure and the ease of airborne dispersion of powders, or the propensity of suspensions to form liquid aerosols.

This section discusses the general principles of prevention and control of worker exposure to free and unbound NP. It is the user's responsibility to decide what means should be applied, depending on the risk assessment in particular situations. Special attention must be paid to nanoparticles that involve substantial or unknown health risks and have little or no solubility in biological fluids. We must also remember that although different studies have shown certain toxic effects in animals after acute exposure, there is practically no knowledge of the chronic risks associated with NP (NIOSH, 2006, 2009b; Ostiguy *et al.*, 2008). In such a situation of uncertainty related to the toxicity of nanomaterials and their exposure level, prudence in limiting occupational exposure is appropriate. SCENIHR (2007) and Maynard (2006) conclude that the risk assessment must be conducted case by case and that the toxicity of new substances cannot be predicted based on current scientific knowledge.

9.1 Prevention

It seems opportune, before discussing how to eliminate or at least reduce and control the risks, to remember that protection of worker health and safety is imperative for corporate development. Production and use of NP can signify different types of risks: toxic products, chemical incompatibility, fire, explosion, electrical risks, high temperatures, etc. It is essential that the senior

management of every establishment make OSH an action priority, especially since the Canadian federal Bill C-45 now holds corporate executives criminally responsible if they fail in their responsibility or act negligently regarding their obligation to protect their workers' health and physical integrity (Department of Justice, 2005). Quebec also has an Act respecting occupational health and safety and various regulations that must be followed. The employer controls the management and supervision of the employees, equipment and work methods. The employer thus has an obligation to adopt all reasonable means to ensure that its employees work in safety. In this regard, it is essential to establish the responsibilities of the individuals in the company clearly and ensure that each person performs specific mandates. This approach also applies to universities and non-university research centres.

The development and implementation of a prevention program specific to the establishment or the research laboratory thus will have to cover different aspects of the question, including identification and assessment of the risks specific to the NP used or produced, assessment of the occupational exposure levels, the criteria and procedures allowing installation of engineering controls, information and training for workers (risks, work procedures, use of equipment, handling of NP, preventive measures and use of personal protective equipment, etc.) and assessment of the performance and effectiveness of the different exposure control strategies. The form of material (powder, suspension or inclusion in a matrix) and the known specific risks (toxicity, high reactivity, flammability, explosivity) must also be integrated into the prevention program, particularly based on the study of the suppliers' Material Safety Data Sheets, the information contained in the literature and the information accumulated in the work environment.

Remember that ongoing information and training for workers represent an essential step in the implementation and maintenance of preventive measures and good work methods. Relying on experts, particularly occupational hygienists can largely help establish a complete and effective prevention program, adapted to the work environment. In Québec, the CSST, the joint sector-based associations, the prevention mutuals, certain consultants, the local health and social services network development agencies and the local health and social services centres (CSSS) can support the development of a prevention program.

The prevention program established should be evaluated and improved regularly to integrate new scientific knowledge and the new elements to be implemented (or the elements already implemented but which must be improved when they do not respond adequately to the stated objectives). Given the wide range of situations that can arise in research laboratories and in companies, the specificities of the prevention program should be adapted case by case (Ostiguy *et al.*, 2009).

9.2 Toxic Risk Assessment

The toxic risk assessment could be considered the estimate of the potential health effects which could result from uncontrolled exposure to certain chemical substances.

Here are the elements, step by step, of a classic health and safety management approach:

- Identify the hazard:
  - Characterization of the particles (physical shape, appearance, diameter, specific surface, surface properties, solubility in water, chemical composition);
  - Emissions (production volume, flow of materials, potential leak);
  - Health effects (animal and human studies) according the potential exposure routes;
  - Environmental effects (persistence, bioaccumulation, transportation over long distances).
- Characterize the risk:
  - Epidemiological studies of the workers, the exposed populations and the consumers;
  - *In vivo* studies: acute, chronic, of different species, exposure routes;
  - *In vitro* studies: different types of cells, models (lung, skin, systemic, etc.).
- Assess the risk:
  - Potential exposure routes depending on the processes: inhalation, cutaneous, ingestion, implementation, parenteral, etc.;
  - Environmental assessment and biological availability;
  - Occupational exposure assessment and means of control in place;
  - Fire and exposure risk assessment.
- Prevent and control the risk;
- Evaluate the effectiveness of the control measures.

A clear distinction must be made between risk and hazard. Hazard is a property inherent in a substance or a situation with the potential to cause effects when an organism, a system or a population is exposed to this agent. Risk is the probability that effects will occur in an organism, a system or a population in specific circumstances (Illing, 2006).

Risk assessment, the process by which the risk is estimated, assumes a good knowledge of the identity of the hazard and the toxicity of the products (dose-response relationship), exposure levels and characterization of the hazards at the various workstations (Herber *et al.,* 2001; Illing 2006; Knight 2006; Ostiguy *et al.* 2009). Integration of all this information and comparison of the dangerous properties and the estimated effective dose based on the exposure levels allows an assessment of the risk level (Illing 2006; Knight 2006). In the case of NP, the previous sections shed light on the major deficiencies in knowledge related to several of these aspects. In the absence of adequate toxicological data on NP, it remains possible to document the known risks for the same substance of a greater size. This should provide minimal information on the potential toxicity of NP. Figure 13 summarizes the information and knowledge normally necessary for risk assessment where:

$$\text{Risk}_{\text{toxic}} = \text{Toxicity} \times \text{Exposure}$$

It must be noted that, for each and every element to be quantified, the current data are insufficient to assign a number to the significance of this parameter.

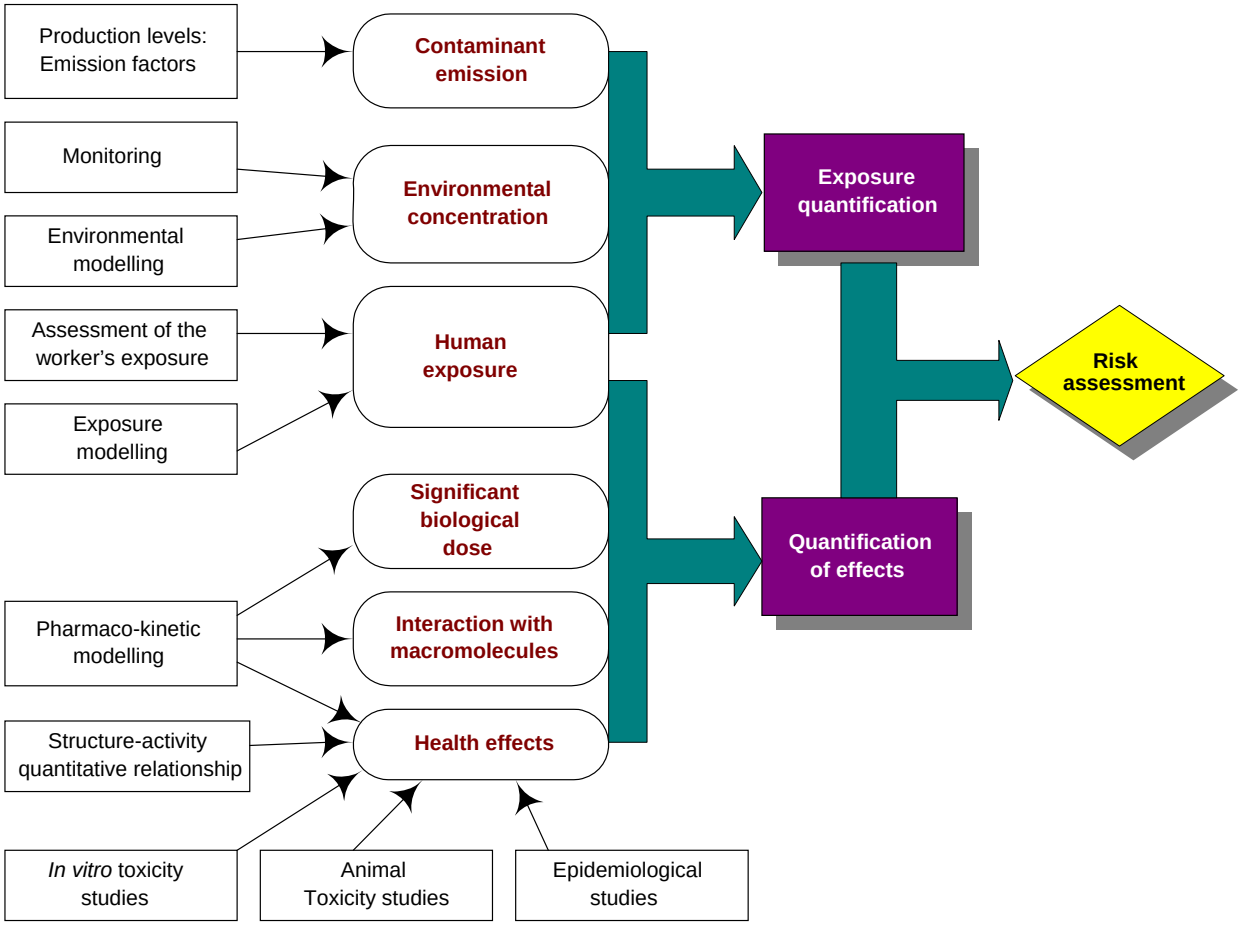


Figure 13 : Classic Approach to Risk Assessment

Thus, it clearly appears that with the substantial limitations of current knowledge, quantitative assessment of the risks related to NP poses a special challenge and will lead to a **high level of uncertainty** in many situations (Kandlikar *et al.*, 2007; Ostiguy *et al.*, 2009). As in the case of any chemical substance, the risk assessment uncertainty models are based on three categories:

- the uncertainty related to chemical and physical characterization, including selection of the best NP characterization parameters, the characteristics of the particles that can affect toxicity, their evolution and their transport in the organism or in the environment;
- the uncertainty related to the dose and the health effects for various exposure routes, the representative parameters that should be measured, the translocation mechanisms to the different parts of the organism;
- the uncertainty related to the lack of knowledge of toxicity mechanisms and disease development.

In each of these fields, there are currently competing hypotheses and models, on which scientific consensus is not yet established. We are not talking here about parametric uncertainties to be included in a model, but uncertainties regarding fundamental choices of the causal mechanisms themselves and the variables that must be introduced in the model. Nonetheless, some authors, including Kuempel (2006a, 2006b), propose human risk assessment models based on extrapolation of data in rats. In default of the ability to apply a quantitative risk assessment model to all NP and in the absence of epidemiological studies, the logical alternative is to apply the judgment of experts or groups of experts in the field and to look for the most plausible trends (Kandlikar *et al.* 2007).

Among these trends, we find that NP toxicity probably can be correlated better to the total surface, the biologically active surface area, the number and size of the particles, and the reactive properties of particle surfaces rather than their total mass. Thus, the danger of NP must be identified according to the special characteristics of the particle in question, namely the type of particle, the size, the structure, its properties and its chemical structure (Tsuji *et al.,* 2006). On the other hand, each assessment will be specific to a particular type of NP and not exportable to other NP, not even to NP of the same type produced under different conditions. As already discussed in detail in Chapter 6, the results of many studies show that, at identical mass, NP often show greater toxicity than larger particles with the same chemical composition. The Material Safety Data Sheets (MSDS) may also be available for certain NP, but in view of the lack of current scientific knowledge specific to NP, the risks and preventive measures identified in these MSDS must be interpreted very prudently. In most situations, the MSDS of the majority of NP indicate the same properties and the restrictions as for bulk materials (Colvin, 2003a).

Risk assessment includes the exposure assessment and the toxicity assessment, which must be studied for the different particles in question, depending on whether or not a dose-response relationship is present. In some cases, volatility, carcinogenicity, flammability, toxicity and persistence in the environment are taken into account for the risk analysis studies (Ogilvie Robichaud *et al.,* 2005). The proportion of respirable particles which can separate from the NP agglomerates must also be considered (Lam *et al.,* 2006).

Faced with multiple unknowns and given that it seems impossible to estimate a Lowest Observable Adverse Effect Level (LOAEL) or a No Observable Adverse Effect Level (NOAEL), it appears preferable, as a precaution, to minimize occupational exposure in order to prevent the risk of overexposure and the development of occupational diseases. Indeed, the precautionary principle, currently recommended by many countries and OHS research institutes, is based on two general criteria (Vineis, 2005):

- an appropriate action should be taken to respond to limited, plausible and credible evidence of a substantial risk;
- the burden of proof is reversed, from showing the presence of a risk to proving the absence of risk.

Moreover, in safety matters, the clouds of organic and metallic NP or of any combustible substance will offer major catalytic, fire or explosive potential. This fact is mainly related to the large reactive surface areas presented by NP. Consequently, the fire or explosive potential must always be considered (Pritchard, 2004).

The optimum risk reduction approach consists of implementing effective control strategies while accounting for the differences between NP and larger particles. We must not forget that individual NP behave like airborne vapours and can travel great distances without sedimentation. On the contrary, agglomerated particles behave more like solid aerosols and airborne dispersion and sedimentation depend on the agglomeration level and ambient conditions. The control strategies may include many elements studied and selected for their adaptability to the treated situation (Roberge *et al.*, 2004).

Moreover, with use of a greater quantity of inert gases, there is a risk of asphyxia in certain processes involving production of NP or their incorporation into value-added products. There is also a risk of electrocution in high voltage and high current processes.

A committee of international experts recently came to a conclusion about the evaluation of the risk of NP (SCENIHR, 2009). This excellent document reviews the main data from the scientific literature, and summarizes the scope and limitations of our current knowledge. Thus, it clearly shows that many hazards have been demonstrated for different synthetic NP. Of these, the toxicity of several NP has been shown, for the environment as well as for humans. However, it should be remembered that not all NP induce toxic effects. As examples, titanium dioxide and carbon black have been used for a long time and have shown only slight toxicity. The hypothesis that the smaller a particle is, the more toxic it is, is not supported by current scientific knowledge. Along these lines, NP are comparable to other chemical products: some are toxic, and others are not. In this matter, a case-by-case approach is therefore recommended (SCENIHR, 2009). Furthermore, recent studies propose results of risk assessment for different substances: titanium dioxide (NIOSH, 2005; Hanai *et al.*, 2009), CNT (Kobayashi *et al.*, 2009) and fullerene C_{60} (Shinohara *et al.*, 2009a).

9.3 The Control Banding Approach

While quantitative risk assessment is impossible at this time in most situations involving NP, a new approach of European design, known as control banding, is emerging in North America. Developed for small and medium businesses, this concept is useful to determine the control levels to be established for substances or mixtures of substances for which the toxic risks are not well known (AIHA, 2007). Thus, the approach is based on exposure control in the absence of standards or sufficient knowledge of toxicity. Paik *et al.* (2008) report they have developed a control banding tool applicable to nanoparticles and have used this approach successfully several times in laboratories. More recently, Zalk *et al.* (2010) also used this approach. The application of this approach is described in detail in the best practices guide (Ostiguy *et al.* 2009) and will not be recapitulated here. Different international organizations including ISO (TC229, WG7) are currently working on the development of a standard based on this approach, while NIOSH recently published a review on the potential of using this approach in North America (NIOSH, 2009c).

9.4 Risk Control and Nanoparticle Control Strategies

In general, there are five main approaches to controlling UFP risks and exposure: elimination, substitution, engineering techniques, administrative means and personal protective means and equipment. These complementary approaches developed for UFP can serve advantageously as a

starting point for NP. The information that served to produce this chapter and the best practices guide mainly comes from expert documents from various countries, including: Ménard, 2004; Ostiguy *et al.*, 2006b, 2009; Aitken *et al.*, 2004; Bruschi and Thomas, 2006; NIOSH, 2006, 2009b; ICON, 2006; ISO, 2007, 2008b; Pritchard, 2004; Bégin and Gérin, 2002; Gérin and Bégin, 2004; US Department of Energy, 2007; BAUA, 2007; Ellenbecker, 2007; Harford *et al.*, 2007; AFSSET, 2008.

While the means of control are relatively well known for UFP, such as welding fumes, control of NP exposure faces a special challenge, namely that the clientele targeted by this control is not necessarily familiar with UFP control technologies. The people potentially at the greatest risk, at least in Québec, are currently found in research laboratories or in businesses newly created to synthesize these products, or users of these products, such as the textile and plastic sectors. Some of these work environments have no experience controlling UFP emissions.

The leading risk control approaches are illustrated in Figure 14. In fact, to minimize workers' exposure risks, the control measures should be considered during design of a research laboratory's facilities or industrial processes, activities and even workstations (Ménard, 2004; Ostiguy *et al.*, 2009). It is the responsibility of the designer and the principal to recognize the risk factors specific to the processes and production modes and plan how to eliminate or at least reduce these risk factors. They must also design and recommend control measures and ensure the effectiveness of the measures put in place (Ménard, 2004).

Given the current absence of standards and the many uncertainties related to toxicity and the safety aspects of NP (fires, explosions), as discussed in the section on risk assessment, it is recommended that a precautionary approach be adopted to NP production, handling, storage, transportation and use control plans. For all situations in which risk assessment cannot be effective based on current scientific research, it is suggested that an ALARA (*as low as reasonably achievable*) approach be used to reduce exposure to a minimum concentration. In the future, when the risks are better discerned and better understood, it will be possible, in some specific situations, to opt for preventive measures that are less strict. But it must never be forgotten that the risks are related to many factors, among which chemical composition is a key element. We have already seen that NP composition can vary infinitely. Also, the processes used for their development, production and use can be very different. Finally, the knowledge relating to the hazards following occupational exposure remains fragmentary. In such a context, the uncertainty relating to the risks associated with the many particular situations found in the workplace should be managed until the magnitude of the occupational exposures and risks is better documented. What is required is a structured prevention approach for identifying the hazards presented by NP, the evaluation of their health, safety and environmental risks in relation to the processes used and the particular conditions of the work methods, and the implementation of effective measures for controlling these risks. The importance of regularly verifying the effectiveness of the implemented preventive measures must be emphasized. Each situation thus will require a distinct analysis.

Since the different risk control approaches were discussed in detail in the best practices guide (Ostiguy *et al.*, 2009), the reader interested in the description of each of these approaches and the assessment of their effectiveness is invited to read the guide available on the IRSST's website (http://www.irsst.qc.ca/files/documents/PubIRSST/R-599.pdf). The parts dealing with engineering control aspects and administrative measures will be discussed very briefly.

In general, approaches that achieve a dust contamination as low as possible must be favoured. These approaches include, for example, the use of automated sealed systems, the use of NP in the liquid phase rather than the solid phase, the collection of emissions at source, filtering of the air before its recirculation or release into the general environment, and the use of personal protective equipment as needed. Training and informing workers, and preventive maintenance of equipment and workplaces represent a few of the many administrative measures necessary for the sound management of NP exposure control.

Although personal protection is the last aspect to be considered, the information provided in the guide is limited. In such a context, more detailed information is presented in this document.

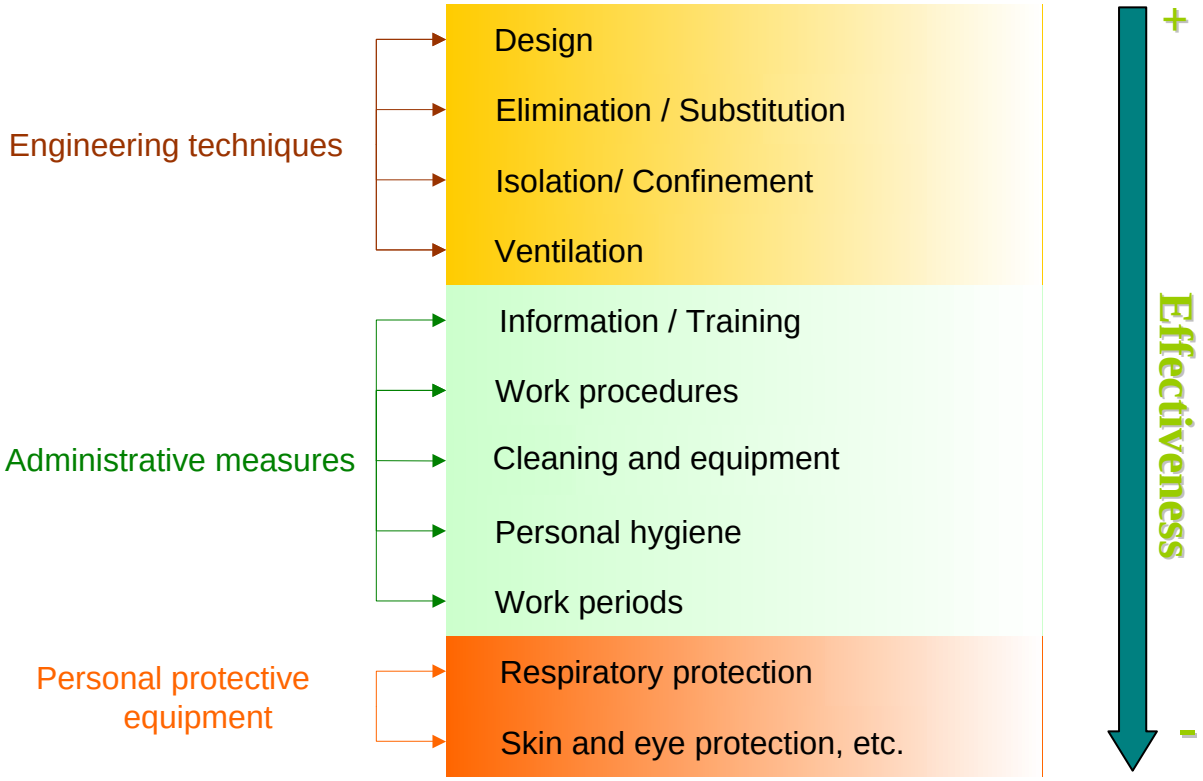


Figure 14 : Risk Control Hierarchy Applied to NP

The effectiveness of these different approaches to exposure control has already been proven with ultrafine particles. A document summarizing current knowledge in this field was recently produced by Safe Work Australia (2009a). Overall, the effectiveness of the usual different approaches is confirmed. Nevertheless, in a context where it is often impossible to quantitatively assess the risk as well as the occupational exposure, it is recommended that a precautionary approach be used, as suggested in the IRSST's good practices guide (Ostiguy *et al.*, 2009; http://www.irsst.qc.ca/files/documents/PubIRSST/R-599.pdf). The authors also raise concerns relating to the quality and the accuracy of labelling and the content of safety data sheets, which could incorrectly orient the choice of means of protection.

More specifically, the report (Safe Work Australia, 2009a) emphasizes that elimination is impossible, since the use of NP is essential in the development of new products. Substitution (product, equipment, work method) is not yet widely used. However, the modification of nanomaterials (fullerenes, CNT, quantum dots, metals and metal oxides) has reduced the toxicity of some products. Current evidence is that properly designed enclosure and confinement substantially reduce or totally eliminate the workers' exposure to the NP that can be aerosolized. These methods, just like many others, are normally used with administrative measures and PPE. Ventilation at source as well as filtration with HEPA filters can significantly reduce or totally eliminate exposure to NP.

9.4.1 Personal Protection

As already mentioned, personal protective equipment is normally used as a last resort, and only when all others means of control have been implemented without being able to protect the worker adequately. Personal protection cannot claim to be a substitute for engineering techniques and administrative means when they do not protect the worker adequately. On the other hand, it may prove to be essential, particularly in handling powders, in equipment maintenance operations or when installation of engineering measures is not completed.

The exposure assessment instruments and techniques described in Chapter 7 can help us determine the effectiveness of the means of protection put in place and the necessity of personal protection. We should note that there are currently no NP-specific standards, even though standards exist for several of these chemical substances in larger dimensions. Moreover, since quantitative risk assessment is often impossible, the decisions on wearing respiratory protection equipment, gloves and protective clothings must often be based on a qualitative risk assessment.

We must not forget that, in general, the risk assessment studies show that, at equal mass, NP can be substantially more toxic than the same particles of larger dimensions. In the guide published in 2007, the U.S. Department of Energy (U.S. DOE, 2007) recommends wearing standard wet chemical laboratory equipment: closed shoes, long pants without cuffs, long-sleeved shirts and lab coats. Several situations will also require the wearing of respirators.

A recent study by the IOM (2009) carried out for the British HSE reviewed the wearing of respirators. The conclusions of this review can be applied to all PPE. Several factors influence the use of PPE and their effectiveness: knowledge about the hazard, perception of the risk, the perceived effectiveness of the protection, the attitudes of upper management and supervisors, and the prevention culture of the company and the workers are the main ones. The researchers (IOM, 2009) also identified some factors that limit the appropriate wearing of PPE. Personal health problems can increase sensitivity to the risk or even make the wearing of PPE more difficult, and a lack of training, respiratory resistance caused by a respirator, a lack of thermal comfort, or discomfort related to pressure, difficulty communicating, and the incompatibility of PPE (making task performance more difficult) represent the main obstacles to the efficient use of personal protective equipment by workers.

It is also clear that management is responsible for the implementation of personal protection programs. Managers must:

- Recognize the importance of prevention and the protection of workers' health;
- Recognize the requirements for personal protection;
- Ensure that a program for PPE is developed;
- Accept their role in making the program effective;
- Ensure that the appropriate equipment is chosen;
- Ensure that the equipment is available in sufficient quantity;
- Play their role to ensure effective implementation and supervision, mainly by setting an example;
- Plan for appropriate cleaning, maintenance and storage.

Several aspects must also be considered regarding the workers, namely:

- A good knowledge of the hazards and an appropriate perception of the risks;
- Involvement of the worker and responsible behaviour;
- The application of the recommendations of the PPE program.

Respiratory protection

In situations where it is necessary to wear respiratory protection, the Quebec Regulation respecting occupational health and safety (2007) makes it mandatory to develop and implement a respiratory protection program.

According to Lara *et al.* (2010), the main elements of a respiratory protection program are:

- Training personnel in the risk, the protection levels, maintenance and storage of respirators, use of respirators according to the manufacturer's recommendations;
- If possible, assessment of their protection factor[4];
- Fitting tests;
- Environmental control;
- Written procedures (for selection, use, training and test performance) related to the different aspects of this program.

This program, approved by the company's upper management, also requires the appointment of a program administrator and assessment of the program's effectiveness, at least once a year, with the objective of ensuring that it is applied adequately by all respirator users.

Depending on the desired respiratory protection level, several classes of respirator exist, which offer different degrees of protection, provided that the worker uses them properly. Table 9 summarizes the assigned protection factors (APF) associated with different respirators by OSHA (USACHPPM, 2006). However, we must not forget that the main limits of respiratory protection most often are the partial seal between the mask and the skin, discomfort and suboptimal

[4] The protection factor (PF) is defined as the quantitative measurement of the seal adjustment of a specific respirator for a specific person. This is the ratio of concentration of a contaminant present in the work environment to its concentration in the air inhaled by the respirator wearer. The assigned protection factor (APF) expresses a protective safety index offered by the respirator. The higher this factor, the greater the protection (*Lara and Vennes,* 2003).

maintenance of the equipment, much more than the filtration efficiency. Therefore it is necessary to distinguish the often substantial variance between the protection factor (PF), which is the protection coefficient in a real work situation, and the assigned protection factor obtained under ideal laboratory conditions. In this way, Grinshpun *et al.* (2009) demonstrated, on a group of 25 subjects, that for particles from 30 to 1000 nm, the particles that penetrate the respiratory system are mainly due to an inadequate seal between the skin and the respirator. Thus, for N95, between seven times more particles (40 nm) and 20 times more particles (1000 nm) entered as the result of an inadequate seal compared to the particles entering through the filtering medium (Grinshpun *et al.,* 2009).

A complete guide to respirator selection and use, produced by the IRSST, is available on the following websites: www.irsst.qc.ca/fr/_publicationirsst_673.html and www.prot.resp.csst.qc.ca.

Table 9 : Comparison of Assigned Protection Factors of Respirators (USACHPPM 55-011-1106)

Type of respirator	OSHA29CFR 1910.134 (2006)	NIOSH (2004)	ANSI Z88.2 (1992) [e]	ANSI Z88.2 (working document)
Air-purifying respirator – quarter mask	5	5	10	5
Air-purifying respirator – filter	10	10	10	5
Air-purifying respirator – adjusted half-facepiece	10	10	10	10
Air-purifying respirator – adjusted full facepiece (if filter ≠ N-P-R 100)	50	10	100	50 [d]
Air-purifying respirator – adjusted full facepiece (if filter = N-P-R 100)	50	50	100	50 [d]
Powered air-purifying respirator – adjusted half-facepiece	50	50	50	50
Powered air-purifying respirator – adjusted full facepiece	1000	50	1000 b	1000
Powered air-purifying respirator – helmet	25/1000 [a]	25	1000 b	1000
Powered air-purifying respirator – loose-fitting facepiece	25	25	25	25
Supplied-air respirator (SAR)– on demand - half-mask	10	10	10	- - -
Supplied-air respirator (SAR)– on demand - full mask	50	50	100	- - -
Supplied-air respirator (SAR)– continuous flow - half-mask	50	50	50	250
Supplied-air respirator (SAR)– continuous flow – full mask	1000	50	1000	1000
Supplied-air respirator (SAR)– continuous flow – helmet	25/1000 [a]	25	1000	1000
Supplied-air respirator (SAR)– continuous flow – ?????	25	25	25	25
Positive pressure SCBA with supplied air-line – half-mask	50	1000	50	250
Positive pressure SCBA with supplied air-line– full mask	1000	2000	1000	1000
Demand open circuit SCBA – half-mask	10	- - -	10	- - -
Demand open circuit SCBA – full mask	50	50	100	- - -

Type of respirator	OSHA29CFR 1910.134 (2006)	NIOSH (2004)	ANSI Z88.2 (1992) [e]	ANSI Z88.2 (working document)
Demand open circuit SCBA – helmet	50	- - -	- - -	- - -
Positive pressure SCBA*– full mask	10,000	10,000	10,000 c	10,000 [c]
Positive pressure SCBA*– mask	10,000	- -	- - -	10,000 [c]
Combined air-line SCBA with auxiliary positive pressure open-circuit full mask	10,000	- - -	- - -	10,000 [c]

SAR: Supplied-air respirator

*: Positive-pressure SCBA often identified as SCBA (Self Contained Breathing Apparatus).

a: The employer must obtain certification from the manufacturer that the device has a protection level of 1000 or more.

b: For HEPA filter if used for protection against particles; if non-HEPA filter, APF = 100.

c: For emergency only.

d: With quantitative fit test (QNFT), or else APF = 10 with qualitative fit test (QFT).

e: Repealed in 2003.

It is also possible to consult the NIOSH document (*NIOSH Respirator Selection Logic*) at the following address: www.cdc.gov/niosh/docs/2005-100/default.html. The information from the NIOSH site are also available in French on the IRSST web site.

Four parameters normally contribute to the efficiency of the air purification elements of a particulate filter, chemical cartridges and filter canisters: interception, electrostatic attraction, diffusion and gravitational sedimentation (Chen *et al.* 2006b) (See Figure 15). Electrostatic attraction is also possible for charged particles because the manufacturing process for certain filters generates static electricity.

It is well known that particles larger than 300 nm are collected on the filters mainly by phenomena of inertial impaction, interception and gravitational deposition, while the smallest particles are collected mainly by diffusion and electrostatic attraction.

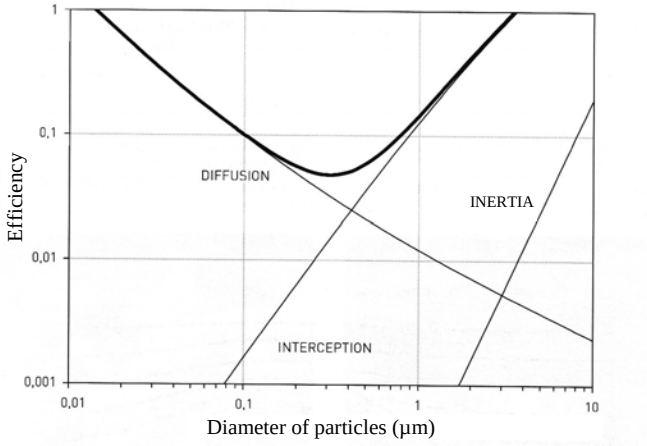


Figure 15 : Parameters Contributing to the Efficiency of Air Purification Elements

This situation means that the 300 nm particles should be the ones normally captured with the least efficiency, thus making them the most penetrating (Hinds, 1999). For particles bigger or smaller

than 300 nm, filter efficiency should increase rapidly. Thus, for example, the efficiency of filtration due to diffusion increases as particle size decreases. Moreover, on the nanometric scale (<100 nm), diffusion is the dominant filtration mechanism (Chen *et al.,* 2006a). This trend continues until the particle is so small (a few nm) that it almost behaves like a vapour. When the particle becomes so small, it is theoretically possible that filtration efficiency could decrease and that the particle would rebound from the fibre without being held by the Van der Waals force. The current studies show, for particles larger than 2.5 nm, that filtration follows the theoretical models and that no thermal rebound is observed experimentally, while filtration efficiency continues to increase down to 2.5 nm, which represents the smallest particle measured (Wang *et al.,* 2007; Kim *et al.,* 2007; Heim *et al.,* 2005; Pui and Kim, 2006). Thermal rebound was observed by Kim *et al.* (2006) for particles smaller than 2 nm.

Wang *et al.* (2007), Japuntich *et al.* (2007), Pui and Kim (2006), Thomas *et al.,* (2008) and Kim *et al.,* (2007) also established that the filtration efficiency of nanoparticles follows the theoretical Brownian capture models resulting in reduced penetration as nanoparticle diameter decreases. Bémer *et al.,* (2006) report that the efficiency of charged filters increases when the charge is high and the gas speed is low. These variations in filtration efficiency also have a significant impact on the dimensions of the most penetrating particles. Thus, Bémer *et al.,* (2006) concluded that the most penetrating particles for an electrically charged filter are 50-60 nm instead of 300 nm for an electrically neutral filter and that, in addition, the filter-particle adhesion, temperature, air flow, pressure and clogging status of the filtration layer alter a filter's efficiency.

However, laboratory experiments have shown that the most penetrating size may vary according to the type of filtering material used, the air flow used and the general conditions of the respirators. Thus, VanOsdell *et al.* (1990) and Dhaniyala *et al.* (1999) have established that, for HEPA filters, the most penetrating particles could range between 100 and 300 nm, while for other researchers, they range respectively between 50 and 100 nm (Martin and Moyer, 2000; Richardson *et al.,* 2006) or between 30 and 70 nm (Balazy *et al.,* 2006; Chen *et al.,* 2006a; Eninger *et al.,* 2008a, 2008b) or between 50 and 60 nm (Bémer *et al.,* 2006). Beyond these diameters, the filtration efficiency increases due to diffusion of the particles striking the filtration material. In another study, Rengasamy *et al.* (2009) show that the most penetrating monodispersed particles range between 30 and 60 nm and are captured mainly by an electrostatic mechanism, with all filters showing "electret" characteristics. Pretreatment of these filters with isopropanol to eliminate static electricity accumulated during the manufacturing process shifted the minimum filter efficiency between 200 and 300 nm. All of the eight filters tested conformed to the expected performance.

Rengasamy *et al.* (2008) evaluated the efficiency of filtration of different approved N95 and P100 filters with sodium chloride (NaCl) and silver NP of different granulometries from 4 to 30 nm in diameter. All the filters conform to the expected performance levels but the nature of the NP, NaCl or silver, influences the efficiency of the filters. Balazy *et al.,* (2006) and Grinshspun (2006) performed tests to validate the use of N95 respirators on mannequins when the respirator was sealed and airtightness was ensured. Certain tests by Balazy (2006) were performed with sodium chloride aerosols in a chamber simulating the working conditions of an indoor environment, at flow rates of 30 L/min and 85 L/min. The other two studies used MS2 bacteriophages. The authors conclude that, as a general rule, the use of N95 respirators conform to their APF except under certain conditions, when the penetration rate exceeds 5%. Indeed, depending on the workers' respiratory rate, the size of the NP and the type of respirator used, the retention efficiency may be

less than 95%. The results obtained showed that when the respiratory rate is at its highest value (85 L/min), the penetration is greater than that obtained at a flow rate of 30 L/min. Also, for a respirator whose filters are charged with static electricity, the penetration rate is greater than 5% for particles ranging in size from 30 to 70 nm, while for uncharged filters, the maximum penetration is around 300 nm (Balazy *et al.,* 2006; Grinshspun 2006).

These outcomes are compatible with those of Bémer *et al.* (2006) for filtration systems. The percentage penetration by the particles would increase in proportion to the respiratory flow. At a flow of 85 L/min, penetration thus would be greater than at 30 L/min (Balazy *et al.,* 2006; Richardson *et al.,* 2006). The evaluated filters were composed of charged polypropylene fibres (electret medium) (Balazy *et al.,* 2006). In the Balazy study, the proportion of N95 filters not conforming to the expected performance levels was as high as 90% at a flow of 85 L/min. The increased quantity of particles in the filter and the leaks caused by a lack of tightness can also increase NP penetration. The lack of tightness can be explained, in particular, by the quantity of particles accumulated in the filter, increasing the loss of charge (Balazy *et al.,* 2006). The Balazy filters consisted of three filtration layers. One filter had three polypropylene layers while the other contained two polypropylene layers and one inner polyethylene layer. For the neutral filters, the maximum penetration was obtained at 300 nm. This fact leads to the suggestion that, whenever possible, uncharged filters should be used to minimize exposure to NP under 100 nm. The electrostatic charge comes from the material used and the manufacturing process.

Richardson *et al.* (2006) also tested N95 and P100 filters and conclude that the penetration rate exceeds the NIOSH requirements (<0.03% for the P100 filters and < 5% for the N95 filters) at high flow conditions for the majority of the filters tested. Penetration rates of up to 20% were measured at high flow with N95 filters and 50 nm particles. The efficiency of a charged filter would increase when the charge is high and the gas speed is low (Bémer *et al.,* 2006). Like Lavoie *et al.* (2007) for microorganisms, Balazy *et al.,* (2006) and Grinshspun (2006) do not recommend wearing surgical masks, because their laboratory tests showed penetration rates between 20.5% and 84.5% for 80 nm NP at a flow of 85 L/min.

It seems important to recall that the practical efficiency of respirators is not only linked to the APF but that other factors have a direct impact on this efficiency in the work environment. Thus, the tightness of the interface between the respirator and the skin is essential to prevent the direct passage of dust. Leaks in this interface can be linked to improper adjustment, poor choice of respirator for the worker's physionomy, duration, type of work to be performed or filter charging. User comfort and maintenance represent key factors that favour good performance of respiratory protection.

Often the practical efficiency of respiratory protection in the work environment is more related to acceptability by the worker, tightness, comfort, work methods and respirator maintenance than to the filtration medium's performance. This is why the use of a positive pressure self-contained breathing unit (SCBA) with a facepiece, instead of type N, P or R, 95, 99 or 100 disposable respirators, will increase the comfort level and the level of protection, particularly by ensuring positive pressure inside the mask, which will minimize the negative impact related to an imperfect fit between the respirator and the skin.

In most situations, unless contraindicated after a detailed analysis of the risks in the work environment, wearing an SAR respirator with P100 cartridge should offer adequate protection. If it is impossible to perform a quantitative risk analysis, the IRSST recommends considering NP dusts to be highly toxic and favours wearing an SAR respirator with very high-performance filters for all potential exposure situations. In situations where this level of protection is still insufficient or there is an immediate risk to the worker's health or life, supplied-air respirators or self-contained breathing apparatuses allow maximum protection.

Cutaneous protection

The development of new knowledge in industrial hygiene is increasing our awareness of the importance of taking into account cutaneous absorption in aggregate exposure risk assessment. The nature of industrial nanoparticle synthesis processes is such that there is a strong probability of cutaneous exposure during the production, handling and use stages or from surface contamination during maintenance and repair of equipment. The product recovery and packaging stages, and general maintenance of workplaces and equipment, all provide opportunities for contact with the skin. Some products that make up nanoparticles can penetrate the skin by dissolution. However, it seems that even certain insoluble products that make up nanoparticles could penetrate the epidermis and possibly end up in the bloodstream, where they can travel throughout the body. Chapter 6 on health risks shed light on the insufficiency of knowledge in this field. Despite the fact that there is currently no cutaneous protection standard, it is preferable, as a precaution, to introduce controls that minimize cutaneous exposure.

Schneider *et al.* (1999, 2000) proved that there are many factors that contribute to the inefficiency of skin protection equipment. The main factors limiting this equipment's efficiency include, notably: (i) direct penetration of solid material or permeation of a liquid through the materials making up the equipment, and (ii) the transfer of substances through direct contact between the equipment surfaces and the skin. Penetration of the equipment by nanoparticles is likely to be even greater than by larger particles, for which tests have shown a high penetration rate. The characteristics of the different NP will influence the penetration rate in the protective equipment, which would be greater in stitches, zippers, sleeves or extremities (Mark, 2005a; Bureau of National Affairs, 2004). The literature does not currently allow the actual efficiency of such equipment to be determined. Because of the small diameters of NP, it is very likely that the effectiveness of some of this equipment is very limited.

Maintenance of skin protection equipment is also an important aspect to consider. Given the context, and as a precaution, it would be desirable, when possible, to use disposable clothing, which normally provides excellent skin protection. For example, the use of Tyvek® hooded coveralls, aprons and shoe covers may provide good skin protection, though the information currently available cannot guarantee absolute effectiveness. The same principle applies to gloves, which are available in a wide range of sizes and resistances to various chemicals, cuts and perforations. The BSI (2008) recalls that selection of gloves must account for the risk and the conditions of use. Gloves must be well adjusted and consider the ergonomic requirements and the user's specific health conditions. Finally, they should prevent exposure without increasing the overall risk. As in the case of respirators, a glove management program should be implemented. This accounts for the tasks, exposure, and selection of the gloves, ergonomics, training of users and foremen, maintenance and safe disposal methods.

Until we can have NP-specific cutaneous protection equipment, it is possible to use the equipment designed for other substances, on condition that certain modifications are made to it (for example, reduce static electricity of clothing to avoid attracting NP) (Bureau of National Affairs, 2004). The Massachusetts Institute of Technology (MIT, 2006) gives an example of nitrile gloves as a cutaneous protection device, which could be used for handling during short-term contact. For longer handling, two pairs of gloves can be worn, one over the other. The U.S. DOE (2007) recommends wearing gloves when NP and particles are handled in a solution and selecting them so that they resist NP and the solvent used. Changing the gloves regularly is recommended to minimize the exposure risk. The contaminated gloves must be put in sealed bags and stored in the hood until their disposal according to the regulations in force. Wash the hands and arms thoroughly after wearing gloves should always be part of the cutaneous protection program.

Since the list of existing means of control is relatively imposing, it is suggested that readers seeking more information consult an industrial hygienist or refer to recent manuals on occupational hygiene, processes, chemical engineering or ventilation. Only a few means of control have been described briefly in this chapter. The efficiency of cleaning and decontamination of cutaneous protection equipment poses a real challenge. It is therefore recommended, whenever possible, to use disposable equipment, which will be discarded safely, as previously discussed. Gloves should be used in all cases of NP maintenance or decontamination work.

Ocular protection

For ocular protection, it is recommended that closed safety glasses, safety eyewear or face shields be worn. One company reports a ban on wearing contact lenses in its laboratories (ICON, 2006). The use of a respirator with a full facepiece allows respiratory and ocular protection simultaneously, as well the possibility of wearing corrective or contact lenses.

Ingestion prevention

Ingestion in the work environment normally results from direct transfer during hand-to-mouth contact. In the case of dust particles such as NP, a portion of the NP accumulated in the upper respiratory system will be transferred to the digestive system by the mucociliary elevator.

Medical suveillance

Currently, the body of scientific knowledge and medical evidence is insufficient to be able to recommend specific medical screening, and thus have NP-exposed asymptomatic workers undergo specific medical examinations (Nasterlack *et al.*, 2008). In this context, the Department of Health and Human Services - NIOSH (2007, 2009a) recently published an interim discussion and improvement guide specifically on medical screening in workers potentially exposed to NP. Even though the evidence is currently insufficient to recommend specific monitoring of workers, it is possible to consider screening in specific situations. For example, if specific medical detection exists for the same substance in larger dimensions, this could also apply to NP. We should mainly consider all existing knowledge relating to biological exposure monitoring of the workers, which could, in some situations, allow a response to be detected by the body and lead to a certain level of vigilance.

For work environments where workers are potentially exposed to NP, these authors recommend:

- Taking measures to control workers' exposure to NP;
- Monitoring the risks based on implementation of control measures;
- Considering the establishment of medical surveillance approaches to help assess the effectiveness of the means of control and identify the health effects and the new or unrecognized problems related to occupational exposure.

The main elements of medical surveillance include an initial medical examination coupled with documentation of each employee's medical and occupational history and periodic medical examinations at regular intervals, including specific medical detection tests. As needed, depending on the test results, more frequent detailed medical examinations could be recommended or do biological exposure monitoring; in the case of higher than normal exposure, after an uncontrolled spill, for example, post-incident examinations and medical screening could be performed. When the risk becomes well documented, a written medical report must be produced; the workers' training then allows them to recognize the symptoms better. After identification of specific risks, the employer must take action immediately to control them. Nasterlack *et al.* (2008) propose general medical screening with methods targeted to certain potential effects and also recommend keeping exposure registers so that large-scale epidemiological studies can be conducted eventually.

9.5 Prevention of Fires, Explosions and other Risks

Beyond toxicity and health risks, NP production can represent fire or explosion risks related to the catalytic effects of these substances (Pritchard, 2004; Kirby, 2005). Many chemical processes are catalyzed by small quantities of substances and catalytic efficiency normally depends on the catalyzing agent's surface area, composition and structure. Nanomaterials have large surfaces, which play a catalyzing role and thus can lead to rapid, even violent and explosive reactions. In processes using large quantities of inert gases, the risks of asphyxia and electrocution related to the use of high voltage and high current methods are possible (Shakesheff, 2005). Some carbon compounds or metal dusts represent substances which are easily explosive or flammable (MIT, 2006). A summary discussion of these aspects is covered in the best practices guide (Ostiguy *et al.*, 2009) but will be developed more fully in this document.

9.5.1 Risks of Catalytic Reactions, Explosion and Fire

Section 6.3 describes the risks of catalytic reactions, explosion and fire. When the ideal conditions are combined to produce an explosion, the main explosive or flammable substances are those with a great propensity to catch fire or explode, namely pyrophoric substances, carbon compounds and dusts of several hydrolyzable or oxidizable metals, some non-metallic inorganic substances and organic substances reacting to air, moisture or other substances with which they can come into contact (Pritchard, 2004; MIT, 2006; Biswas and Wu, 2005). Aluminium, magnesium, zirconium and lithium are some examples of substances with high explosive potential (Shakesheff, 2005).

NIOSH (2006, 2009b) affirms that the risks of fire or explosion are greater for a combustible material of nanometric dimensions compared to larger-scaled substances, for the same mass concentration. This phenomenon is explained by the increase in the surface area and potential properties of NP. In fact, reduction of the size of a particle of a combustible compound can reduce the minimum ignition energy and increase the combustion potential and rate (NIOSH, 2006,

2009b). This can induce the possibility that a relatively inert compound will become a combustible material.

9.5.1.1 Key Factors

For a fire to occur, three factors must be present: a combustible material (wood, metal, dust, etc.), a substance or a gas which can cause it to burn (oxygen, peroxide, air, etc.) and an ignition source (heat, flame, spark, reaction between two substances) (INRS, 2005). The last factor is the most conclusive because, in most cases, the first two factors are almost always present. These aspects were already discussed in section 6.3.

9.5.1.2 Risk Characterization

According to Dinyer *et al.* (2005), explosion risks can be characterized by tests performed on the different substances suspected of being explosive. Some must be performed under controlled conditions, by accounting for the particles' size, their concentration in water and the moisture in the air, for example. One of the tests cited by the author is the minimum ignition energy test of a substance, which reveals the minimum energy necessary to cause it to explode. Another is the explosion severity test, which can provide a virtual prediction of the scope of the damage. However, it is not always possible to perform these tests because, due to the cost, the necessary quantity of NP (500 g) is not constantly available (Dinyer *et al.*, 2005). Therefore, these tests are more or less useful for several NP. Some reference methods exist: ASTM E2019-99 (Standard Test Method for Minimum Ignition Energy of a Dust Cloud in Air) and ASTM E1226-00 (Standard Test Method for Pressure and Rate of Pressure Rise for Combustible Dusts (Dinyer *et al.*, 2005). Proust (2005) affirms that analytical methods and techniques exist to characterize explosion risks, but they apply to larger particles.

Granier and Pantoya (2004) proved that a nano-scaled Al/MoO_3 alloy ignites 300 times faster than the same alloy of micrometric size. More recently, Bouillard *et al.* (2008) of INERIS showed that certain aluminium dusts of nanometric dimensions, although previously passivated with air and partially agglomerated, led to an explosion much more violent than those observed with the same product of larger size.

Nano-scaled particles have long been used as catalysts on a large scale. Depending on their composition and their structure, certain nanomaterials thus could trigger catalytic reactions and increase the risk of explosion or fire, which normally would not be anticipated solely from their chemical composition (Pritchard, 2004).

9.5.1.3 Risk Reduction / Explosion Prevention

Explosions can generate enormous pressures and cause damage to workers and physical structures. They can project debris almost everywhere and possibly release particles into the outdoor air. The reduction of these risks depends on their identification and seeking means to prevent them: processes modification, use of more resistant materials, reduction of the number of workers, etc. Kirby (2005) mentions that the explosions, which caused the most losses of human life and damage to equipment and buildings were secondary explosions.

This reduction is possible, but to achieve this goal, it is essential to know the characteristics of the NP used (size of particles, composition, state, minimum activation energy, etc.) and the conditions of the work environment (ambient temperature, space available, etc.). The composition of the particles also requires attention, because they can contain solvents or other substances capable of changing the activation energy necessary to produce an explosion (Pritchard, 2004). These two factors combined – the low spark ignition threshold and the natural production of charges during handling – automatically increase the risk of trigging an explosion by electrostatic sparks. This phenomenon could require the development of special methods to combat it.

The reduction of the explosion risk depends on control of the three components necessary for an explosion – oxygen, the combustible material and the ignition source (flames, heat, friction, etc.). Control of the comburant material (oxygen) involves its elimination or reduction of its concentration in the risky environments, while keeping the workers' safety in mind (especially if they have to enter risky locations). Dinyer *et al.* (2005) affirm that it is possible to prevent explosions with an oxygen concentration of less than 8%. However, they recommend adding a 3% safety factor and maintaining the risky locations at an oxygen concentration of 5% or less by adding other gases such as nitrogen or carbon dioxide. The INRS (2005) also mentions this method for fire prevention, particularly by a reduction of the oxidation reactions with certain substances. Given that it is impossible for workers to work under such conditions, they will have to be applied in closed-circuit processes or in controlled-access storage areas.

It is more difficult to control the combustible material because the dusts present in the environment are usually the raw material. However, a good knowledge of the characteristics of the substances present and their dangers can minimize the risks. Finally, if they are not eliminated completely, the ignition sources must be identified clearly and controlled. For example, it is possible to modify the process, isolate the sources in precise locations, connect and ground them to avoid charging them with electricity, or ensure that the equipment is always in good working order (Dinyer *et al.*, 2005). Installation of electrically insulating materials can help reduce the risk of triggering an explosion by static electricity.

Moreover, methods such as process modification, regular equipment maintenance and capture at source can reduce the number of particles which can accumulate in the environment. Regular maintenance can prevent accumulation and resuspension of particles, particularly by vibration of the pipes. In addition, the storage method must be designed to limit the release of airborne NP, reduce the danger of products mixing in the event of leaks in the containers, and finally, control the environmental conditions (temperature, ventilation, oxygen rate) (Dinyer *et al.*, 2005 and Pritchard, 2004). Kirby (2005) summarizes the main means of prevention as follows:

- prevent dust accumulation outside the equipment;
- use dustproof mechanical and electrical equipment whenever possible;
- prevent dust emissions from open bins and drop points;
- maintain the highest workplace maintenance standards;
- eliminate the ignition sources;
- isolate the risky operations, either by distance or by construction;
- install explosion vents on the equipment and buildings;

- ensure adequate fire protection;
- store these materials in sealed containers or tanks;
- handle the materials in closed and sealed tanks or pipe systems;
- the disposal systems must prevent formation of dust clouds and accumulation in the workplaces and on the work surfaces;
- train the employees in the risks of combustible dusts and the prevention measures.

In case of explosions, more specifically, the consequences can be reduced by installing materials resistant to the released pressures, anti-explosion panels and explosion-resistant containers (Peters, 2005b; Whitaker, 2005). Peters (2005b) and Whitaker (2005) affirm that it is possible to install systems capable of detecting pressure rises and releasing stabilizing products (such as inert powders) when the pressures are too high. Installing an explosion vent is also a method to prefer (Shakesheff, 2005, Whitaker, 2005). Whitaker (2005) adds some protection methods regarding pharmaceutical powder explosions as an antistatic dust collector or a device allowing ventilation outside the collector or neutralization of the effects of the explosion (pressure, dusts, gas and fire due to the explosion). The last method reported by Whitaker (2005), which would only rarely be used, consists of containing the explosion in the dust collector.

9.5.1.4 Risk Reduction / Fire Prevention

To prevent a fire from occurring, it is important to identify the dangers properly – i.e., identify and characterize the products likely to produce fires (physical state and physicochemical characteristics). This information should be contained in the Material Safety Data Sheets of the products used and made available to the workers. It is then essential to know the conditions of storage and use of the substances: temperature, volume, type and tightness of the containers, ventilation, access control, separation of the products, lighting, construction materials, etc. (INRS, 2005). The environmental conditions where the substances are handled should also be known, as well as the quantities used, transported or stored and the identification of the potential fire sources (or activation sources): thermal (heat sources), electrical (sparks, heat release), electrostatic (sparks), mechanical (heat), climatic (lightning, sunlight) and chemical (reactions with heat release) (INRS, 2005).

Maintenance of apparatus and systems can avoid production of heat or sparks originating fires. The analysis of possible effects of an equipment operating error can also indicate potential fire sources.

Fire prevention involves several stages. Above all, risks must be avoided. If this is impossible, it is essential to try to control them. Replacement of combustible products (often impossible with nanoparticles) with products of little or no combustibility can prevent the outbreak of a fire. The oxygen concentration can also be reduced by replacing it with an inert gas, but it is then essential to pay attention to the fact that workers may need to enter the oxygen-deficient locations. Precautions for their safety must be taken. It is also necessary to analyze the different processes to detect places where a fire can start and its probable consequences (breakage, collapse, explosion). Once the inflammation sources are identified, it is imperative to establish the means of reducing the risks: for example, by installing a cooling process for heat sources, by optimizing the means of control, by adding detectors or instituting regular maintenance and inspection of machines and pipes. It is also possible to add certain procedures or modify the equipment to make it safer.

The layout of the sites and the construction materials used must allow limitation of the potential effects of a fire: materials chosen according to their fire resistance or their reaction to fire, obstacles to propagation of the fire by addition of firewalls, isolation of risky premises. It is also essential to ensure that the exits are easily accessible and equipped with alarms and that the premises are accessible to rescuers. Detectors (smoke or temperature) must be installed. Finally, it is indispensable that the means of detection and extinction are in good condition, that the emergency exists are clearly identified and that the personnel are trained for emergencies with the protocols to follow in case of disaster.

The extinguishers present in the workplaces must be chosen according to the types of materials used and the risks related to their use. But other means can also be used. Shakesheff (2005) suggests putting powders that are burning or on the verge of burning in a steel container capable of reducing the oxygen concentration and ensuring their transport to a safer place. Whitaker (2005) adds that it is possible to use rapid closing valves or chemical barriers with inert powders.

Fire prevention should account for the regulations in force, particularly concerning electrical installations. Given the very low granulometry of NP and their very long sedimentation time, the electrical equipment should be protected against dusts, and even be vapour-tight in some cases. Additional precautions should also be taken regarding their operating temperature and the increased risks of self-ignition of NP.

The consequences of a fire can concern humans (burns, asphyxia, poisoning, crushing by structures and stress) as well as materials (destructions) and buildings (collapses). The substances used to extinguish the fire are also capable of causing damage to buildings and the environment. Finally, the costs may be high. Thus, it is more advantageous to prevent fires than to repair the damage.

In the scientific literature, no NP-specific documentation is found regarding firefighting. However, the principles that generally apply to pulverulent materials should be adopted for NP, with special caution for easily oxidizable metallic dusts.

The choice of extinguishing product will consider the compatibility or incompatibility of the material with water (Ostiguy *et al.,* 2006b). The metallic dusts react with water to form, among other products, hydrogen, which ignites easily and deflagrates. Chemical powders are available to extinguish metallic dust fires. During a metallic dust fire extinguishing operation, it is essential to ensure not to create major air movements that would have the effect of putting the metallic dust into suspension and thus increasing the risk of deflagration. Finally, to reduce the risks, it may be necessary to use a controlled-atmosphere production and storage process (carbon dioxide, nitrogen or inert gas).

9.5.1.5 Catalytic Reactions

Another NP-related risk is the possibility of triggering catalytic reactions or violent reactions, which can be very dangerous (Pritchard, 2004). Catalytic reactions depend on the composition and structure of NP (NIOSH, 2006, 2009b). For example, NP and porous materials of nanometric dimensions have been used as catalysts for years to speed up reactions or reduce the temperature necessary for reactions in liquids or gases (NIOSH, 2006, 2009b).

9.5.2 Storage

Protection of the properties of products and control of the risks related to the reactivity of certain particles are specific aspects of nanomaterial storage. The tanks must be very tight to prevent leaks and contamination of the sites (dispersion, slow sedimentation) and to allow different NP reactivity and granulometric characteristics to be taken into account. Layouts similar to those used for gas storage must be considered.

Storing nanoparticles also involves special protection to conserve the products. The small size of the particles, which often tend to agglomerate, provides a very large surface area in contact with the surrounding air, thereby facilitating chemical reactivity. To protect against loss of NP-specific properties, oxidation, and even explosion in the case of certain metals, different preventive procedures will avoid any deterioration of the product and any risk of fire or explosion. Among the possible solutions is storage in the presence of an inert gas or under anhydrous conditions so as to protect certain metals from oxidation and even explosion. In other situations, NP can be coated with a protective coating composed of salts or different polymers, which can then be eliminated before the product is used.

9.5.3 Routine Maintenance and Workplaces

Regular workplace maintenance is necessary to eliminate dust on the floors, work surfaces, equipment and walls and avoid the risk of airborne resuspension or explosion. The equipment should be cleaned thoroughly, simultaneously using appropriate administrative measures such as padlocking; this preventive maintenance will minimize the risk of emergency interruption of production and will ensure a safer operation.

The workplaces should be cleaned at the end of each shift, using a vacuum cleaner with a HEPA filter, installed correctly and replaced regularly, or wet methods according to the specific conditions of the process and the products involved and their respective risks: explosibility, flammability, incompatibility with water, etc. The efficiency of such systems must be evaluated. In some cases, the electrostatic charge of the NP will be taken into account to select the best system. Use of a compressed air spray or a broom which could resuspend dust, should be rejected as in any dry process. In all cases, however, the cleaning procedures must avoid any contact between the worker and the scrap, which will be disposed of in accordance with the laws and regulations in force.

Whenever possible, it is recommended to use wet cleaning methods with soap or cleaning oils. Commercially pre-moistened or electrostatic microfibre wipes can also be used. The wipes and other materials used for cleaning must be disposed of as hazardous waste and not reused to avoid any possibility of resuspension of airborne dusts. Finally, the establishment’s prevention program should determine which personal protective equipment is to be used during cleaning or during product recovery operations in case of spills or accidents.

9.5.4 Spills

Currently there does not seem to be any guide to the procedures to follow in case of an NP spill. Consequently it would be prudent to base our strategies for spills and contaminated surfaces on the existing best practices, while considering the available information on risks and potential exposure routes, depending on the type of NP. Since NP tend to disperse in the air, it is necessary to wear respiratory protection equipment (with a high-efficiency filter) and a cutaneous protection device during handling. Among the usual procedures for liquid or solid spills, and depending on the nature of the NP, we note the use of vacuum cleaners with HEPA filters, humidification of dry powders, use of wet wipes to clean up powders and application of absorbent materials (NIOSH, 2006, 2009b). As in the case of housekeeping, it is absolutely essential to avoid procedures, such as use of an air spray or a broom, which could resuspend dusts. Finally, waste handling and disposal must be performed in accordance with the laws and regulations in force.

9.5.5 Waste Disposal

Any waste disposal from processes using NP should conform to the by-laws, regulations and standards in force at the municipal, provincial and federal levels. Since the real risks to the environment related to NP are not documented in the vast majority of situations, it would be prudent to consider NP, and the objects that served to clean them (wipes, gloves, etc.) as hazardous materials and dispose of them in an environmentally safe manner. Currently, disposal of nanomaterials depends on the type of original substrate in the waste (MIT, 2006). Waste management companies can assist the establishment or the university in this task; others prefer to treat their waste themselves. Among the possible strategies are in-situ chemical treatment of nanomaterials before their disposal, recycling or returning nanomaterials to the suppliers and incineration of organic NP.

9.5.6 Other Aspects of Prevention

We could discuss several other aspects of prevention, but we will not do so here, given their specificity to the products used, synthesized or handled and to the processes implemented. Furthermore, the literature tells us nothing about the specific measures to be put in place with NP. Among others, consider the preventive aspects to be developed and implemented for asphyxia, electrocution, emergency protection equipment and first aid.

10. REGULATION AND RECOMMENDATIONS

One of NT's distinctive features resides in the fact that in many cases, the physical and chemical characteristics of substances, as well as their potential health impacts, may be altered, deliberately or unintentionally, not by changing their chemical composition, but by changing their physical structure or surface properties. In such a context, specifically regulating nanoparticles represents a major challenge. This is why several countries are simultaneously forming expert committees to review current scientific knowledge and advocating the development of voluntary standards coupled together with the application and adaptation, wherever possible, of the regulations currently in force (Bartis and Landree, 2006; Balbus *et al.*, 2006, Mantovani *et al.*, 2009). Also, several major multinational companies, including insurance companies, are very concerned by the risk of prosecution should NP cause health problems (Bartis and Landree, 2006). Thus, several of them are hoping that standards and regulation will be developed. Recently, Mantovani *et al.* (2009) and Hansen (2009) reviewed actions taken across the world to develop NT regulation. The European Community is recommending an incremental approach including various actions and initiatives to develop regulation appropriate to NT. Thus, it recommends supporting research initiatives in health, safety and the environment, promoting risk assessment during the entire product life cycle, defining and implementing appropriate regulation, establishing a dialogue with all actors and developing international coordination (Mantovani *et al.*, 2009). Many countries are now recommending a precautionary approach.

10.1 Necessary Ethical Rules

In the absence of sufficient knowledge on the health risks and effects of exposure to NP, it is essential for employers, workers, investors, lawmakers, health authorities and the general population to provide itself with ethical rules to guide decision making (Schulte and Salamanca-Buentello, 2007; Obadia, 2008; Mantovani *et al.*, 2009; Sander, 2009). Moreover, at the moment several organizations in a number of countries are working out "voluntary standards" with a view to setting up guidelines for the safe and responsible development of NT.

The risks associated with the toxicity and spread of NP, currently impossible to quantify in many situations, dictate a responsible and precautionary stance. Thus, the UN maintains that the international community should adopt rules restricting NT use to avoid possible abuses in fields such as the right to privacy and military uses.

In the case of OSH specifically, a number of issues raise ethical questions. These include risk identification and communication by employers, scientists or authorities, risk acceptance by the employee, selection and monitoring of control methods, medical screening programs and even investing in research on evaluating toxicity and controlling exposure (Schulte and Salamanca-Buentello, 2007).

After the *Conseil Général des Mines* (General Mines Council) and the *Conseil Général des Technologies de l'Information* (General Council on Information Technologies) published a report including 13 recommendations in November 2004, France's CNRS gave its view in October 2006, (Dupuy and Roure, 2004); an NT working group was created as part of the *Comité Consultatif National d'Éthique* (CCNE, National Consultative Ethics Committee for Health and Life

Sciences). In Great Britain, the Royal Society and the Royal Academy of Engineering (2004), and in the Netherlands, the Academy of Arts and Sciences (Levelt, 2004) indicated, amongst other things, the importance of examining NT-related ethical problems. The United States, the EEC and many Asian countries also raised this concern.

In Québec, the *Commission de l'éthique de la science et de la technologie* (Science and Technology Ethics Committee) examined all ethical aspects of NT and nanomaterials and presented a report to the Government of Québec in the autumn of 2006 (*Commission de l'éthique de la science et de la technologie*, 2006). It recommended in particular: a) the application of the precautionary principle in all phases of the product life cycle and inclusion of this principle in governmental policy in order to avoid all harmful health and environmental effects; b) intervention at the federal government level so that monitoring agencies can evaluate the toxicity of nanotechnology products before authorizing their marketing; c) assurances that the ethical research committees are properly equipped and have received the support needed to evaluate research protocols on the use of nanotechnological substances and products in the medical field; d) prior to their marketing, the setting up of a system to track the potential effects of nanotechnological products on the environment, as well as a procedure for withdrawing them in case deleterious effects on the environment are observed; e) the beginnings of a completely open process of information exchange with the population to properly define the scientific, economic, social and ethical issues associated with NT development; f) the creation of a multidisciplinary research program on the impacts of new technologies and risk management; g) the taking into account of NT development in the area of employment and employment training; h) the creation of an NT information portal for the general population.

10.2 Regulation in European Nations and the European Community

It should be noted that by the time NT action plan for 2005-2009 was published (in 2005), the European Community had indicated its commitment to promoting the responsible development of NT. This action plan clearly noted the need to provide a high level of public health, safety, protection for the consumer and the environment, as well as the need to include social dimensions, the development of standards, appropriate regulatory approaches, the application of a code of conduct and international cooperation.

The European parliament (2009) made a series of 37 recommendations to the Commission and the member states. The first recommendation emphasizes that the European parliament "is convinced that the use of nanomaterials should respond to the real needs of citizens and that their benefits should be realised in a safe and responsible manner within a clear regulatory and policy framework (legislative and other provisions) that explicitly addresses existing and expected applications of nanomaterials as well as the very nature of potential health, environmental and safety problems." Among the other sections, it is clear that the European parliament does not legitimize the position of some countries with the sole application of the regulations currently in effect without taking into account the specificities of the risks related to NP. The European parliament (2009) hopes that provisions will be developed that target nanomaterials in a specific way, and invites the Commission to review all the legislation within two years.

Regulation is being revised or questioned in many countries in Europe, Asia and America. For further information, readers may refer to Mantovani *et al* (2009), Hansen (2009), Knebel et Meili (2010) and the following website: http://www.innovationsgesellschaft.ch/nano_regulation.htm. In Europe, NP procedures are currently subject to laws and regulations in force relating to chemical substances. Nevertheless, lawmakers in several countries are clearly calling for the adoption of a precautionary approach to carefully evaluate and balance NT risks and benefits (Hock *et al.,* 2008; Obadia, 2008; Mantovani *et al.*, 2009; NIOSH 2009b; Hansen, 2009).

In conformity with *directive 67/548/CEE (articles R.231-52* and following of France's *Code du travail, arrêté du 20 avril modifié*), the person in charge of marketing substances must perform an evaluation and make a report to the competent authority of the member State involved. For a product that has never before been marketed, this directive applies regardless of the production or import tonnage (even if simplified files are anticipated for the production of less than one ton per year per manufacturer). Thus, the regulation facilitates evaluating the nanomaterial so that is tailored to the actual production and the taking into account of dangers specifically related to its nanostructure form. In light of the requirement to create a safety data sheet for all production over one ton, the evaluation will give rise, among other things, to statutory labelling warning about the dangers of the substance. For substances marketed after 1981, *directive 67/548/CEE (article R.231-52-12* of the *Code du travail)* holds that the notifying party must inform the competent authority about all new knowledge on the health effects of the substance and about its new uses (AFSSET, 2006).

NP from chemical substances must meet the requirements of REACH regulation (Registration, Evaluation, Authorisation and Restriction of Chemicals). This regulatory framework of the European Union came into force in 2007. A key REACH component, based on a precautionary principle, is that the burden of proof regarding their safety and non-harmful effects on health is now on manufacturers, importers and producers, rather than legislators (Mantovani *et al.,* 2009). The principal objective of this new program is to provide a higher level of protection for human health and the environment, while allowing European markets in the chemical industry to function efficiently.

The European obligation to protect production, marketing, transport and use of new materials concerns manufacturers who must determine the properties and potential exposure resulting from the production and use of nanomaterials. Manufacturers will produce a new Derived No Effect Level (DNEL) in the context of REACH and this value will serve as a threshold limit value (TLV) when developing safety data sheets for substances that have no TLV. The European Nanobusiness Association is a body whose mission is to promote professional development for NT industry and trade in Europe. Among the activities of this association are the drafting of standards, providing information to the public, analysing legislation and providing expert testimony for political leaders and decision makers. Several other bodies, both in the pharmaceutical and agri-food industries, have been formed and are evaluating the impact of the introduction of new products (European Nanotechnology Gateway, 2004).

Different approaches are also anticipated in the planning of European regulation. In particular, insurance and reinsurance companies are playing a greater role. The legal repercussions of the effects of asbestos prompted them to make representations to the government to demand the implementation of a regulatory framework for nanomaterials. Two approaches in particular emerge

in this regard: one advocating self-regulation and the introduction of voluntary programs by the "NT" community, and the other recommending the addition of a regulation that would be complementary to the REACH directive in Europe. Other regulations must also be taken into account including those relating to the *Agence medicale européenne*, the European Food Safety Authority, aspects of health and safety in the workplace, and the environment (Mantovani *et al.*, 2009). Until now, the only amendment to be introduced was to cancel the exemption status under REACH for carbon and graphite, since it was considered inadequate in taking into account the potential risks and current uncertainties of carbon-based nanomaterials (Hansen, 2009).

Without being a regulatory organization, the British BSI (2008), basing itself upon knowledge relating to larger sized particles, made specific recommendations for threshold limit values (TLVs) by classifying NP in four major categories:

- Fibrous materials: It is recommended that the TLV currently in effect (0.01 fibres/mL) be applied by doing the counting by electron microscopy and by retaining the usual counting criteria (length greater than 5μ, length/width ratio > 3);
- For carcinogenic, mutagenic or reprotoxic substances, it is recommended that TLVs ten times smaller than those currently in effect be adopted;
- For insoluble nanomaterials, the BSI recommends that the same relationship that NIOSH proposed for titanium dioxide be applied to all insoluble NP, namely to reduce the existing standard by a factor of 15 while retaining a TLV based on the mass of the NP. They also suggest a TLV of 20,000 particles per millilitre while differentiating these particles from the natural background for UFP;
- Finally, for soluble NP, it is proposed that a TLV be applied that represents half of the TLV of a particle with the same chemical composition but of larger dimensions.

This British position, proposed at a meeting of an international ISO committee, has not been retained for now.

10.3 Asia-Pacific Region

Due to its economic and commercial status, Japan is the leader in NT development in the Asia-Pacific region: it accounts for half the total R & D budget in the field. However, the uneven economic status of Asian countries influences the regulatory measures proposed to protect populations and workers. Indeed, the policies of poorer countries, such as India, Pakistan, Sri Lanka and Nepal are not as strict as those of more developed countries, such as Japan, Taiwan and Singapore when it comes to protecting populations and the environment (Lorrain and Raoul, 2004). Of course, NT can provide tremendous benefits to populations. However, they can have potentially devastating effects unless all factors are taken into consideration. Australia considers responsible NT development a priority. They closely follow the development of knowledge in the field and serve on numerous international committees of experts. Also, they consider their current regulations sufficiently robust to deal effectively with NP problems, that is, without having to introduce further regulatory changes (Harford *et al.*, 2007).

10.4 North America

10.4.1 The United States

To the best of our knowledge, there is currently no specific regulation for NT in the United States, except for the law on R & D in NT in the XXI century (Public Law 108-153) promulgated in 2003. New mandates have just been entrusted to various institutions to examine, and if necessary modify, the existing regulation on NP and nanostructures (Marburger and Connaughton, 2007). On the other hand, there are federal regulations on evaluating and regulating the workplace and the environment, as well as on the health risks of materials and new technologies. An October 2005 workshop organized by the Rand Corporation, at the request of NIOSH (Bartis, Landree, 2006), and discussed by Kuzma (2007), revealed four worrisome situations in the area of regulation:

- a lack of knowledge regarding risks, associated with institutional responsibilities, that can slow down the development and marketing of new products;
- efforts to evaluate occupational health hazards linked to specific nanomaterials may be hampered by the lack of scientific knowledge on the major classes of nanomaterials;
- a low level of public and private investment in improving knowledge on OHS and the environmental risks linked to these new materials, compared to the spectacular investments observed in R & D for new products;
- the indispensable collaboration between federal agencies and the public and private sectors.

In November 2007, the Office of Sciences and Technologies Policies and the Council on Environmental Quality (Marburger and Connaughton, 2007), came up with principles to monitor nanomaterial-related aspects of OSH and the environment. They revealed that federal agencies with regulatory functions, including the EPA, the FDA, the OHSA and the NIOSH are responsible for setting up polices for protecting the environment and the health of populations. In addition, agencies directly involved in nanotechnology R & D, or using NT as part of their job, must monitor them properly. The message was fully understood and in an amendment to the Act setting up the National Nanotechnology Initiative (NNI) and approved by the American House of Representatives in June 2008, the NNI was required to coordinate the agencies more effectively so that it could provide the resources required for the ethical, legal and environmental aspects of NT, as well to ensure that other social concerns linked to NT were taken into account (Mantovani *et al.*, 2009).

Among the guiding principles are the need to transparently develop the knowledge required to understand NT impacts, standardize and render coherent the practices of various governmental agencies in the area of risk evaluation and management, encourage international cooperation, support access to and sharing of information, and develop and introduce special laws and regulations whenever a need is identified and supported by appropriate scientific knowledge (Marburger and Connaughton, 2007). Lastly, these laws must promote NT development.

Today, the occupational health institutions to which Americans refer are NIOSH, OSHA, ACGIH and the FDA. OSHA, NIOSH and ASTM select protective equipment according to established criteria, and NIOSH (2007, 2009b) has developed a guide to best practices in the workplace. There is no approval or certification for consumer products (except for food and medicine). The

challenge is daunting because one might have to deal with a chemical product that comes in different forms (diamond, carbon nanotube, graphite, carbon black, fullerene, carbon nanofoam); or with materials in which the only difference is granulometric (such as TiO_2).

The Environmental Protection Agency (EPA) has the authority to regulate nanosubstances via various programs covering the entire product life cycle. These include, among others, the Toxic Substances Control Act (TSCA), the Clean Air Act (CAA), the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA), the Clean Water Act (CWA), the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and the Resource, Conservation and Recovery Act (RCRA). To date, it would seem that the TSCA has been the NT regulation most often referred to by the EPA. In this context, the EPA adopted a new regulation taking effect on August 24, 2009, and dealing with 23 chemical substances, including single- and multi-walled CNT (The Synergist, 2009; USEPA, 2009).

In this context, it is possible that the EPA will view the production of nanomaterials as the production of new chemical substances, with all the preventive measures that this implies, in conformity with the laws and regulations in force. A notice period of 90 days could then be required to allow the EPA to revise information associated with the new substances or with their health, safety and environmental effects (Mantovani *et al.*, 2009) before a manufacturer imports or handles these products. For substances representing an unreasonable risk for human exposure, the EPA requires that workers wear an N-100 respirator and protective clothing (The Synergist, 2009). After study, the EPA could also decide to prohibit or limit the production or use of these substances.

The FIFRA, the Federal Insecticide, Fungicide and Rodenticide Act, was applied in 2007 in a case that became famous. In a controversial decision, the EPA deemed that the Samsung washing machine, which, following the washing of each load, was found to be covered with silver nanoparticles and releasing a certain quantity of silver ions, should be registered as a pesticide. In November 2008, the EPA decided to classify NTC as “new products”. This meant that the latter would have to be approved for use whereas this was not the case before since they had the same chemical composition as graphite, a substance that had been approved for a long time. They had made known their intentions in October 2008 (USEPA, 2008a). The EPA also issued new rules for alumina NP modified by siloxanes (USEPA, 2008b). As previously mentioned, the EPA adopted a new regulation taking effect on August 24, 2009, and dealing with 23 chemical substances, including single- and multi-walled CNT (The Synergist, 2009; USEPA, 2009). The EPA is currently in the process of reviewing the TSCA with possible implications for NP.

OSHA has various standards that are applicable to NT. For example, nanosubstance manufacturers must provide material safety datasheets describing the properties of these materials, their health effects and preventive measures to be implemented. If the monitoring techniques are inadequate, protective respiratory equipment must be provided to the workers. Lastly, the OSHA moral responsibility clause obliges employers to keep the workplace free of recognized hazards. In the workplace, OSHA applies a regulation based on Permissible Exposure Limits (PELs), threshold limit values for chemical substances regulated in the United States that currently have no specific standards for NP (Mantovani *et al.*, 2009). NIOSH recommends threshold limit values that do not have the force of law, as does the ACGIH. As concerns personal protective equipment, OSHA

applies the regulation, the American Society for Testing and Materials (ASTM) determines the standards and NIOSH evaluates and approves the equipment.

With regard to food and drugs, the Food and Drug Administration (FDA) has significant regulative jurisdiction. When, in July 2007, it published the report of a working group on the analysis of NP-specific aspects of science and regulation (FDA, 2007), it had already approved 24 nano-drugs and made note of 26 others in the clinical trials phase. The FDA considered the NP inserted into sun cream as particles of reduced dimensions and not as new particles, and consequently not requiring new approval. The same perspective seems to apply to nanoparticles added to food (Mantovani *et al.*, 2009).

10.4.2 Canada

Since NT constitutes a new field of research, its risks and benefits are continuously being examined and evaluated. Thus, the Government of Canada continues to regulate (Health Canada, Environment Canada, 2007). It recognizes the need to adopt a balanced management approach allowing Canadian society to use NT responsibly. This ensures an integrated and coordinated management of its economic, environmental, ethical, health and social concerns, while preserving the high or improved standards in the areas of safety and the environment (Santé Canada, Environnement Canada, 2007). To this end, Health Canada commissioned the Council of Canadian Academies to set up a committee of national and international experts to determine what is known about the properties of existing nanomaterials, and about their health and environmental effects, and to support the development of regulations focussing on research, risk assessment and monitoring needs (Conseil des académies canadiennes, 2008). This expert committee maintains that current regulatory mechanisms should be strengthened. To ensure worker safety, its members are recommending (a) the development of an interim classification of nanomaterials; (b) a revision of the criteria determining if a new material or product needs to be examined from the standpoint of its health and environmental effects and (c) the perfecting of standardized methods for handling nanomaterials. The authors stress the importance of strengthening metrological approaches to facilitate efficient monitoring of NP impacts on consumers, workers and the environment. They recommend support for an open and enlightened public debate and, to avoid any duplication of effort and the creation of incompatible regulations, emphasise developing a coordinated approach among governmental agencies, as well as with our international partners (Conseil des académies canadiennes, 2008).

Canada's regulatory framework responded to the uncertainties through a precautionary approach giving priority to health and environmental protection. With this in mind, the report of the Council of Canadian Academies (2008) has already set out a partial list (reproduced here) of Canadian regulatory measures that may be relevant in the area of nanotechnology.

Environment Canada

Canadian Environmental Protection Act, R.S.C. 1999, c.33: New Substances Notification Regulations (Chemicals and Polymers), SOR /2005-247 and Persistence and Bioaccumulation Regulations, SOR/2000-107.

Canadian Environmental Assessment Act, R.S.C. 1992, ch.37

Fisheries Act, R.S.C. 1985, c. F-14.

Canada Agricultural Products Act, R.S.C. 1985, c.20.

Feeds Act, R.S. 1985, c. F-9

Fertilizers Act, R.S. 1985, c. F-10

Pest Control Products Act, R.S.C. 1985, c. P-9

Oceans Act, S.C. 1996, c.31

Arctic Waters Pollution Prevention Act, R.S.C. 1985, c. A-12

Canada Water Act, R.S.C. 1985, c.11.

Health Canada

Food and Drugs Act, R.S., c. F-27, Section 1: Food and Drug Regulations, C.R.C. c.870; Medical Devices Regulations, SOR /98-282; Cosmetic Regulations, C.R.C., c.869; Natural Health Products Regulations, SOR /2003-196

Consumer Packaging and Labelling Act, R.S. 1985, c. C-38

Hazardous Products Act, R.S.C. 1985, c. H-3: Controlled Products Regulations, SOR /88-66; Ingredient Disclosure List SOR /88-64

Health of Animals Act, 1990, c.21

Workplace Health and Public Safety Programme: Workplace Hazardous Material Information System (WHMIS).

Canada, Human Resources and Social Development

Canada Labour Code (R.S., 1985, c. L-2), H-3.3: Canada Occupational Health and Safety Regulations, SOR /86-304; Provincial labour codes and OHS codes.

In February 2009, Canada became the first country to legislate on nanomaterials specifically. The law obliges companies and institutions that have produced or imported more than ten kilograms of certain identifiable NP (lithium phosphate, homopolymerized silane) to submit all the information they have on these products: their physical and chemical properties, toxicological data, manufacturing processes and uses. Health Canada and Environment Canada will use this information in the evaluation and management of nanomaterial risks (Gazette du Canada, 2009). The health and environmental departments are currently studying the NTC file and could eventually make specific recommendations on legislation to the Government of Canada.

10.4.3 Québec

In Québec, application of the *Act respecting occupational health and safety* (AOHS) and the *Regulation respecting occupational health and safety* (ROHS) cover general aspects of obligations in terms of developing the workplace contaminant prevention and containment programs of institutions. Several chemical substances contained in nanomaterials are already cited in Appendix I of the ROHS, which clarifies standards for chemical contaminants. However, this regulation does not take into account the granulometry of particles, or the possibility that toxicity may vary according to this granulometry. However, particle shape and granulometry may be of decisive importance in terms of a product's absorption, its distribution and interactions and, consequently, its toxicity.

The Workplace Hazardous Materials Information System (WHMIS) is a Canadian system of which Québec is a member and that obliges suppliers to label chemical substances and produce material safety datasheets describing products; they must also provide their principal characteristics, their health risks and the protective methods they require. Employers must ensure that the MSDS are available and train their workers as to their implications. Several other provincial and federal laws may apply to NP in the same way they apply to chemical substances, as in the case of the transport of hazardous material, for example. However, to our knowledge no Québec law deals specifically with NP.

11. THE MAIN ACTORS IN QUEBEC

Quebec is highly involved in nanotechnology research and is the first Canadian province to have developed an overall strategy to promote the development and commercial production of nanotechnologies. NanoQuebec plays a major role in planning and structuring nanotechnology. It is a non-profit organization co-financed by the Government of Quebec (via the *ministère du Développement économique, de l'Innovation et de l'Exportation*, the *ministère des Affaires municipales et des Régions* and *Valorisation-Recherche Québec*) and the Government of Canada (via Canada Economic Development for Quebec Regions). Its objective is to create critical masses of researchers and the synergy required for improving the research and status of nanotechnologies. It bases its approach on consultation and networking, that is, getting the various actors in the field of nanotechnology to interact (companies, researchers, governments, manufacturers and financiers). All of its initiatives are geared toward eventual business and industrial applications. Concerned by ethical, OHS and environmental questions, it created an Internet portal to circulate information and educate the general population about NT issues (http://nanoquebec.ca/nanoquebec_w/site/index.jsp).

It has already concluded several national and international partnership agreements to help Québec's strategic positioning and maintains several strict partnerships with the main Québec universities (including Université de Sherbrooke, Université Laval, École Polytechnique, McGill University, Concordia University, École de technologie supérieure, Institut national de recherche scientifique and Université de Montréal) and various collèges d'enseignement général et professionnel (CÉGEP, Colleges of General and Vocational Education).

Québec has an extensive university network, with over 50 research groups working in the area of NT. Moreover, it is in research and teaching that we currently find the largest contingent of individuals potentially exposed to NP, though about forty companies, several of which are in the start-up phase, are also involved. To date, it is estimated that there is a total of about 2000 individuals producing or educate about NP, and this figure should increase with the development of new NP products and applications. The proportion of these individuals who are potentially exposed remains undetermined for now.

The IRSST and the CSST are developing a pool of NT expertise to assess existing knowledge on OHS risks. This should help institutions take preventive action, and prevent accidents and increases in occupational disease. To respond to the concerns of their partners, and in their common interest, the IRSST, CSST and NanoQuébec have jointly produced and published a guide to best practices in the workplace (Ostiguy *et al.*, 2009). Also, the IRSST and NanoQuébec recently launched a call for research proposals on various aspects of OHS. Four major projects have been funded and are in progress. Although NanoQuébec has close ties with all actors in this sector, we still do not know the extent of nanomaterial use in Québec, be they produced locally or imported.

12. POTENTIAL AVENUES OF RESEARCH

The NP field is expanding rapidly and the research to date has focused mainly on technological developments for production of new materials. However, the impacts of these materials on health and the environment have only been established partially: this research is in its infancy and there is a manifest lack of scientific knowledge.

Reading hundreds of documents for our literature review nonetheless allowed us to identify several research teams in this field and detect many OSH-related research avenues, which are divided into two sections in this chapter. The first section will offer a general, non-exhaustive vision of the multiple identified or perceived needs. Some of these projects are already being implemented in different countries and could allow further probing of various OSH aspects related to NP. The IRSST eventually will be able to exploit this new knowledge by updating its literature reviews (Ostiguy *et al.*, 2006b, 2008, 2009) and by knowledge brokerage strategies with the organizations possessing information necessary for the IRSST's mission. In the second section, in view of the potential research orientations (only part of which are reported here) and the requirements for developing scientific knowledge, the authors of this report will propose research priorities to the IRSST with the aim of enriching those described in its 2006-2010 strategic plan (IRSST, 2006) and in its 2009-2011 research plan (IRSST, 2009) and of helping to transfer them to Québec work environments and to the safe development of NT in Québec. The IRSST should also assume a provincial coordinating role for research in this field, particularly through its involvement in the Québec occupational safety and health research network community known as the (RRSSTQ - Réseau de recherche en santé et en sécurité du travail du Québec), whose research is oriented to nanotoxicity and covers broader aspects of OSH than those proposed in the second part of this chapter.

12.1 The Main Orientations for Developing Research in the World

A very wide variety of NP are produced, each of which may display its own physical, chemical and biological properties. These NP may follow different penetration routes into the body, some of which are voluntary (drugs, diagnostic tools) and others involuntary (contamination of air, water, soil, food, etc.) or directly related to the environmental conditions of the work environment. Beyond voluntary injection, absorption can occur through the pulmonary, cutaneous or digestive routes. Current knowledge suggests that alteration of their surfaces or of certain of their properties has a direct impact on health effects and the environment, apart from the fact that some properties may also change during the life cycle of products containing NP. The risks related to a specific NP could also evolve over time, as could the means required to control them.

Effective risk management improves when risk assessment is exhaustive. This risk cannot be concretized precisely, given the lack of essential information. For the time being, it is necessary to fall back on precautionary approaches.

Regarding the inevitable needs for new knowledge in OSH, current or future research should aspire to a better understanding of the fundamental parameters for assessing, managing and controlling OSH risks. We will select the following studies, among others, as the main orientations: Mark, 2005a, 2005b; Department for Environment, Food and Rural Affairs, 2005;

DEFRA, 2007; NIOSH, 2008, 2009d; Hurt *et coll.*, 2006; AFSSET 2006; Environmental Defense – Dupont, 2007; Tran, 2006; National Science and Technology Council, 2007; ICON 2008; Conseil des académies canadiennes, 2008; National Nanotechnology Initiative, 2008; Schulte *et coll,*, 2009; Schuster, 2007:

Orientation 1: Harmonize the definitions and produce reference materials

- Several organizations, including the ASTM and ISO, are currently working on the development of a terminology specific to the nanotechnology field. This work must continue and lead to an international consensus.
- The development of well-characterized reference materials (size, granulometric distribution, specific surface, concentration, crystallinity, solubility, chemical composition, electrical charge, morphology, structure, degree of aggregation, mechanical, electrical, magnetic and optical properties, surface chemistry, solubility, etc.) is essential to a better understanding of NP behaviour in the air and in the body. These reference materials must be made available for all the major NP categories (nanotubes, fullerenes, quantum dots, metal oxides, etc.). They will make it easier to compare the results of the future studies, particularly regarding health effects and contribute to the calibration of equipements used in metrology.

Orientation 2: Produce a picture of the Québec situation

- One of the critical stages in risk control is to identify the clientele to be served. This objective will be achieved by developing a detailed inventory of Québec research laboratories and establishments producing NP, as well as establishments incorporating NP into their end products. The challenge quite obviously will consist of keeping this inventory up to date and identifying the establishments that purchase NP, and incorporating them into their production processes.
- In addition to inventorying, classifying and categorizing NP by type, volumes of production or use and applications, extract the relevant data for Québec, including the number of potentially exposed workers.
- Determine the characteristics of materials/products produced/used (size, concentrations, volumes, composition, structure, morphology, surface properties, explosive properties, etc.) and their main uses.
- Determine the main processes implemented in Québec and identify potential associated risks.
- Document the current working conditions, the means of prevention and their efficiency and the good work methods already in place.
- Establish a watch function to keep the data up to date.

Orientation 3: Document the toxicity of different NP

- Unlike several gaseous, liquid or solid materials, the desirable properties of NP are closely dependent on the dimensions, shape and physical or chemical structure of the material employed and their unique properties at these dimensions. Similarly, it is likely that biological activity is closely related to the physicochemical parameters of NP, thus leading us to

reconsider the classical interpretation mode of toxicological studies. The current results of the toxicological research on CNT, fullerenes, quantum dots, silica, titanium dioxide and other oxides and metals that are insoluble or only slightly soluble reveal the need for painstaking examination of methods for characterizing nanomaterials during assessment of their potential biological activity. The classical interpretation of the observed effects directly correlated to mass no longer facilitates understanding and explanation of the observed phenomena. However, a better parameter (or series of parameters), which can link exposure to the observed effects, has not yet been defined. In such a context, it will be appropriate to pay attention to the most complete characterization possible of the NP studied.

- The use of reference materials should be advocated to facilitate comparison of the results of different studies, given that the current studies suggest that absorption, distribution, metabolism, excretion and toxicity depend on the physical and chemical characteristics of NP, their functionalization, their coating, their dimensions and their degree of aggregation. The often divergent results of various toxicity studies of the same products could be attributable, at least in part, to the use of products with the same chemical composition but having different properties. The often very summary characterization of the initial products cannot ensure that the different teams really used equivalent products.

- Given the number and the very great diversity of the NP produced, continued development of new approaches in toxicological research is suggested. The goal is to analyze these products while meeting deadlines at reasonable costs, and to produce a first assessment and a summary classification of toxicity. The normalization of the methods for detection and study of the biological effects of NP would allow establishment of strict and reproducible methodologies, the results of which would be comparable from one study to the next.

- Characterize the NP that will be put on the market, based on their reactivity and their interaction with human tissues, so that categories can be established: inert NP, toxic NP by direct interaction, or toxic NP by indirect interaction, such as through the release of toxic metallic ions. Toxicological research would benefit from the perfecting of new ultrasensitive dosing methods to detect ultratraces.

- For each type of NP, document their capacity for penetration and absorption through physiological barriers (intestines, lungs, skin, hematoencephalic barrier, placenta, cell, cell nucleus), their translocation and the intercellular and intracellular transportation mechanisms and their distribution in the organism, the molecular, cellular and tissular interaction mechanisms between NP and the organisms, their biopersistence in the cells and tissues, the elimination mechanisms (e.g.: how the NP are deposited and eliminated in the pulmonary routes after intratracheal instillation, pharyngeal aspiration or inhalation exposure), their transformation, generation of reactive oxygen species, induction of oxidative cellular stress causing inflammation, followed by fibrosis and various pulmonary impairments, mutagenicity, genotoxicity and cancerogenicity, cell damage (cytotoxicity and apoptosis) and their circulatory effects. The epidemiological studies of UFP suggest specific study of the impact of NP on the cardiovascular system and the brain after pulmonary absorption.

- Determine not only the distribution in the various organs but also bioaccumulation, biodegradation, biopersistence, location, biological fate and toxicity, for each target organ, of the different types of NP in various types of animals following acute and chronic exposure via representative routes of occupational exposure (inhalation exposure and cutaneous exposure).

- Document thoroughly the potential impact of these pulmonary or cutaneous exposures on various types of NP in order to establish dose-response relationships that will facilitate development of exposure limits in the work environment. This can be achieved by measuring, among other factors, the nature and severity of lung damage, oxidative stress, inflammation and fibrosis after pulmonary exposure to NP of different compositions, forms and granulometries.

- For substances displaying toxicity, it is essential to determine the physical parameter best correlated to the toxic effects measured: product mass, number of particles, size or specific surface, charge, crystallinity, etc. It also seems important to approach the study of the effects on model organisms of chronic absorption at low doses. Among the biological species to be studied, some authors recommend paying special attention to those species for which the genome has been sequenced and is accessible, and to genetic study to benefit from the considerable advances of the genomics approach (functional genomics, transcriptomics, proteomics, metabolomics).

- Determine whether certain NP can achieve a transplacental passage and, if necessary, document the eventual consequences.

- Determine whether NP exposure has effects on the immune system, the central nervous system or the reproductive system, or shows cancerigenic or genotoxic effects.

- Determine how the shape, durability and chemical composition of NP affect their translocation capacity and their biological activity.

- Determine whether *in vitro* tests are predictive of *in vivo* effects and how *in vivo* tests on animals can be extrapolated to humans, knowing, for example, that rats are much more dust-sensitive than humans.

- Understand how chemical and physical modifications influence the toxicity of various types of NP and establish the relationships between the structure and the activity.

- Modelling the integration of various complexity scales (cells, organs, organisms, individual, population) could eventually contribute to a better understanding of NP toxicity, once the different parameters essential to application in the models have been determined experimentally and the best practices retained. These models would allow description of the relationship between the internal NP dose and the biological response and determine the minimum doses that may cause potential human health effects.

Orientation 4: Development of risk assessment models and standards

As discussed above, quantitative assessment of NP-related risks is currently difficult, or even impossible, for the vast majority of NP. To the best of our knowledge, NIOSH (2005a) is the only body that has succeeded in producing such an assessment, and only in the case of titanium dioxide. However, recent studies allow new questions to be raised about these conclusions (Warheit *et al.*, 2006, 2007, 2009). In this context, once essential information becomes available, we can envision research on quantitative assessment of NP-related risks.

- Develop statistical methods to consolidate the risk estimates for a certain number of plausible dose-response models.

- Investigate the application of risk assessment methods by using existing data as a base to develop risk management strategies.

- Ultimately, the toxicological studies should make it possible to understand the action mechanisms of different types of NP and establish dose-response relationships capable of estimating safe exposure thresholds and standards for workers. They thus should establish the measurable parameters of NP in the work environment, capable of relating the exposure doses to the health effects.

Orientation 5: Determine the risks related to explosions and fires

- Understand and determine the chemical and physical characteristics and properties of NP in a state to trigger catalytic reactions, and thus to increase the fire or explosion potential and develop methods for their assessment.

- Determine the dustiness related to different NP in the laboratory by normalized methods.

Orientation 6: Verifying safe work thresholds through epidemiological studies

The Québec researchers working in nanotechnology research have been clearly identified but we have little knowledge of the composition of their teams. No information regarding the companies and workers incorporating NP into value-added product manufacturing is currently available, while information on those producing NP is only very partial. In such a context, the following research avenues can be envisioned:

- Conduct longitudinal epidemiological studies immediately, particularly among the cohorts of young technologists being trained in NT. This would make it possible to determine the "referential" health status of a population that could become exposed to NP, with each worker potentially becoming his own control. However, this approach has many limits: limited cohort, unspecified current and future NP exposure, totally unknown future health effects, very high costs.

- With increased NP production and use, conduct workplace epidemiological studies of users and producers to isolate the measurable specific risks in the exposed workers. It will be difficult to cover this aspect for many years, at least in Québec.

Orientation 7: Develop occupational exposure measuring devices and strategies

A research priority should be assigned to the understanding, prediction and quantification of the physical and chemical properties and behaviour of NP in the air and in the body. In the short term, it is necessary to have devices for detecting synthetic NP that can be used in an industrial setting, in the environment and in toxicological studies. These devices should have sufficient sensitivity in terms of size and concentration, good reliability and a low cost, be simple to use and have a spaced maintenance frequency with the possibility of computer networking of surveillance. All this should allow description and measurement of exposure levels with adequate parameters. However, there are no devices adapted to the current industrial context. More specifically, but without limitation, it is therefore proposed to:

- Develop new equipment accompanied by adapted strategies allowing specific measurement of the NP exposure of workers and researchers, in relation to the measuring parameter(s) best adapted to product toxicity assessment for all types of NP, whether particulate or fibrous. The development of new methods for using existing techniques will allow characterization of the relevant parameters of nanoparticulate aerosols with regard to the potential health effects. The selection of well-characterized standards is recommended to develop reference protocols for measuring the atmosphere in the work environment or in the generation chamber.

- Instruments are also required to monitor and characterize NP in toxicological studies and in the body.

- Currently, several types of equipment exist that allow non-specific measurement of different parameters (size, surface area, number of particles, etc.) in the laboratory and, with more difficulty, in the work environment. They are normally bulky, heavy and not NP-specific, lack sensitivity to particle size and the minimum measurable quantity, and often necessitate very specialized expertise. Due to the lack of equipment that can be worn by the worker in the respiratory zone, it is impossible to determine the specific surface of particles, the number of particles, the size, the surface properties, the electrostatic charges or the airborne granulometric distribution at the personal workstation in the work environment. Consequently, new portable equipment will have to be developed to allow better aerosol characterization, particularly by measuring the concentration expressed in specific surface and giving information on the number of particles, the granulometric distribution, the composition and the charge of the particles. Ideally, these instruments should be able to discriminate between NP and other airborne contaminants. Finally, we should note that NP characterization will be facilitated by the availability sought for reference products capable of allowing adequate calibration of the instruments.

- Ideally, develop more sensitive detection methods, specific to the engineered nanoparticles, so as to free the readings from the high and fluctuating background noise of the UFP already present. For example, these methods should be capable of detecting the constituent material or a specific optical property of the particles of interest.

- Until these ideal instruments exist, apply assessment strategies based on the best knowledge available in seeking to characterize a maximum number of airborne NP parameters and understand their airborne behaviour, including their agglomeration, displacement, sedimentation and chemical and physical transformations in the atmosphere.

- Develop and construct NP aerosol generation chambers allowing laboratory reproduction of the anticipated concentration levels representative of work atmospheres in terms of granulometry, concentrations and other characteristic parameters, such as agglomeration state, form, density, electrical charge, chemical composition, etc. These enclosures should be capable, in particular, of studying the behaviour of the different NP in controlled environments containing NP, either alone or in the presence of other contaminants. They should also measure different parameters simultaneously with different equipment; this will allow calibrating the equipment and determining its performance. Finally, we should note that these enclosures are necessary to expose animals through inhalation in a controlled environment.

- Develop knowledge of the chemical characteristics of NP for a better estimate of exposures and risks. The development of chemical analysis methods coupled with sampling methods will often constitute the only approach allowing a distinction between the NP and UFP present in

any work environment. These methods will have to account for the low concentration levels expected for certain NP and the presence in the work environment of other particles of nanometric dimensions (UFP), which can interfere with the other equipment that will make determinations not specific to NP (surface area, number, charge, etc.).

- Develop measuring devices and methods capable of continuous assessment of the work environments and individual exposures and optimize the existing equipment to make it perform sufficiently for assessment of NP.

- Develop specific techniques for each type of manufactured nanoparticle to lower the detection limits and increase the specificity of the analysis.

Orientation 8: Assess occupational exposure

In general, the occupational exposure assessment will be based on the description of the Québec situation (Orientation 2), on the specific toxicity of NP (orientation 3) and on the availability of strategies and equipment allowing representative and distinct measurement of NP exposure (Orientation 7). Since most of the synthesis processes will be performed in closed or partially closed circuit, a dysfunction or leaks on the equipments are always possible. It is then probable that the events originating in airborne NP emissions are fleeting or unstable, or come from specific situations in the process (bagging, etc.), inducing spatial and temporal variability of the concentrations, the granulometry and possibly other parameters, such as electrostatic charges. On the other hand, certain users or processes can operate in an open atmosphere. This would be the case particularly in the textile industry, where suspensions containing NP would be vaporized directly on fabrics. Given the operators' mobility, multiple emission conditions and air movements, the fixed-station data obtained cannot be transposed directly into personal exposure data without special consideration, as is the case for the majority of occupational exposures, independently of the contaminant. It is thus appropriate to conduct certain research to assess occupational exposure to the various NP encountered in industrial establishments.

- Develop occupational exposure assessment strategies, depending on the products, the processes involved and the work situations encountered in Québec.

- Determine the most appropriate measuring parameters for quantifying the risks (surface area, number of particles, surface reactivity, etc.) and assess the performance and precision of the different measuring and characterization instruments (ideally continuously and in real time) and the intervention strategies in establishments to quantify these parameters.

- Develop testing and assessment systems allowing comparison and validation of the sampling and measuring instruments for different NP of various sizes and shapes, including assessment of CNT, particularly by the use of laboratory characterization methods.

- Measure and characterize the NP emission sources and the behaviour and fate of NP under real airborne conditions in the work environments, including dispersion, transformation, evolution, persistence, sedimentation and resuspension.

- Document and characterize occupational exposure (pulmonary, cutaneous and oral route) in various work environments and for different synthesis and use processes. These studies include experimental validation of the deposits in each part of the lung.

- In the toxicity assessment plans, account for the development of worker exposure measurement and assessment tools and the tendency of NP to agglomerate.
- Document exposure during the manufacturing, use, recycling and elimination stages, and during spills or accidents.
- Determine, under real conditions in the work environment, the dispersion, agglomeration, sedimentation and resuspension of NP. With certain powders, a laboratory or in-plant study of the propensity to form an aerosol in response to agitation, a spill or an air flow is of great interest to the industry and industrial hygienists.
- Model and validate the NP dispersion and aggregation models in the work environments.
- Based on the results obtained in the previous stages, determine an overall strategy for monitoring the processes, workers in industrial establishments and research laboratories, particularly in terms of the type of measurement (personal or fixed station), the parameters to be measured (surface area, number, granulometric distribution, etc.), the equipment to be used and the analyses to be performed in the laboratory, the frequency and duration of measurements, etc.

Orientation 9: Assess the efficiency of the means of control

Since various toxic effects have been documented for a wide variety of NP, worker exposure and dispersion of NP in the work environment should be minimized. In this sense, the IRSST, in collaboration with the CSST and NanoQuébec (Ostiguy *et al.*, 2009) have produced a best practices guide to support companies and research laboratories in the implementation of measures for working safely with NP. However, current knowledge on the efficiency of various means of prevention regarding NP is extremely limited. Several aspects need to be documented quickly, for different types of NP of different sizes, dimensions and forms. For example, the first publications on the efficiency of high-performance filters in blocking NP are very encouraging, at least as concerns tested spherical or quasispherical particles. Filter behaviour and efficiency are only one of the topics for which additional research is required to assess the real efficiency of the means of collective and individual protection currently available. Finally, access to new scientific knowledge will allow better assessment and better management of the NP exposure risk. Until then, however, the general principles of occupational hygiene described above allow implementation of the basic elements of sound risk management. In this sense, and non-exhaustively, research on assessment of the efficiency of the means of control should:

- Assess the efficiency of the means of prevention already used in establishments:
  - Design: elimination or substitution of products, processes and equipment…
  - Engineering controls: isolation and enclosure, ventilation at source, local ventilation, ventilation by dilution, laboratory hoods, glove boxes, air recirculation and filtration, equipment isolated against explosions…
  - Administrative means: prevention program, reduction of work periods, safe work procedures, access reserved to authorized personnel, reduction in the number of workers, modification of work practices, personal hygiene measures, housekeeping and preventive maintenance of equipment, information and training for workers, storage, emergency showers and double locker rooms, spill control and management…
  - Personal protection: variety of equipment used.

- Study the explosivity characteristics of NP and the best means of safe production, storage, transportation and implementation.
- In the existing systems, assess the efficiency of all the control measures commonly used during production, bagging, handling, storage and transportation of NP and their integration into value-added products.
- In the laboratory, assess the efficiency of the personal protective equipment available (gloves, aprons, clothing laboratory coveralls, etc.).
- In the laboratory, assess the efficiency of the different filtration systems and existing types of filters, both for the ventilation systems and for the respirators at different representative flow rates for their use by the workers.
- Cutaneous protection could necessitate the development of specific equipment and adapted strategies, the performance of which will have to be assessed.
- Assess the efficiency of the conventional means and determine the best approaches to cleaning of the contaminated surfaces. Also, study the state of the deposited nanoparticle, its granulometry, its adhesion to the collector surface, the ease in dislodging it from the surface and the best cleaning and decontamination strategies, depending on the type of NP.
- Assess under real conditions all the systems and strategies assessed in the laboratory.
- Based on the acquired knowledge, refine the best practices for handling of nanomaterials.
- Conduct research to improve the performances of existing equipment where the assessment shows that equipment performance and procedures are insufficient to lower exposure to an acceptable risk level.

Orientation 10: Promote the safe growth of Québec businesses and safeguard the health of researchers and workers

Existing knowledge on NP and research on the nine preceding orientations has provided a vast array of usable information to favour safe development of NT. Thus, a best practices guide addressing both the work environment and the research environment was produced (Ostiguy *et al.*, 2008, 2009) with the aim of supporting the development of Québec NT. On the operational level, we must:

- Update this guide when new scientific and operational knowledge permit this.
- Develop dissemination strategies specific to these clienteles, most of which are very small production businesses or small research teams. For example:
  - Continue collaboration with the CSST and NanoQuébec, which will participate actively in the dissemination of information to their clienteles.
  - Send a copy of the guide to every establishment or research laboratory in the nanotechnology field.
  - Make all the documents available on the IRSST website.
  - Inform Québec's OSH stakeholders, particularly the CSST inspectors, physicians, nurses, hygienists and hygiene technicians of the occupational health network, the parity sector associations and the prevention mutuals that can act as relays to the various clienteles concerned.

- Multiply the presentations at special interest events (NanoQuébec conferences, for example) or general interest events (regional CSST' conferences, conferences of hygienists, physicians, OSH managers in establishments or universities, etc.) and within the context of training for future nanotechnologists, OSH specialists and engineers, etc.
- Publish the research results in peer-reviewed specialized journals.
- Popularize the research results in articles addressing the various clienteles concerned.
- Produce technical data sheets when this is possible and relevant.
- When the information becomes available, inform the regulatory review committee of the need to adopt NP-specific regulation and tools allowing application of the regulation in OSH.
- Consider the possibility of producing a website based on examples of best practices implemented in laboratories or establishments.

➢ As already mentioned, updating the three documents (Ostiguy *et al.*, 2008, 2009 and 2010) will favour optimum use of new Quebec and international scientific knowledge.

12.2 The Main Research Development Avenues proposed to the IRSST

The previous section showed the enormous need to develop new scientific knowledge relating to the OHS aspects of NP. With limited human and economic resources, the IRSST cannot hope to cover all of these scientifically interesting aspects. Many toxicological studies are currently in progress in the international scientific community and cover a wide range of NP. Simply keeping track of the publications appearing in this field is a challenge. Very few studies are related to occupational exposure. However, since the risk associated with NP is not only related to the toxicity of these substances but also to the workers' exposure level, the authors of this report propose that the IRSST channel its research efforts to the following four avenues and monitor the scientific literature for the other research orientations described above:

o First, continue incorporating new knowledge from world research, particularly but not exclusively concerning toxicology and health risks. This should cover all the orientations described above.

o Second, assume a strong provincial leadership and coordinating role through the RRSSTQ. In particular, this network facilitates collaboration on numerous research projects and funding from various granting bodies in niches that the IRSST would not necessarily be able to fund, either because their relevance and priority have not been recognized, or because resources are too limited. The particularly targeted priorities would cover Orientations 3, 4, 5 and 6.

o Third, establish its own research priorities and orientations to ensure that it has the necessary knowledge for its mission. To this end, ensure that the work directly subsidized by the IRSST pertains primarily to improving knowledge of the Québec work environments where NP may expose workers to greater risks, assessing occupational exposures to these NP, and undertaking programs of prevention (more specifically regarding the effectiveness of means of prevention). The request for research proposals issued by the IRSST and NanoQuébec in autumn 2008 takes this very direction: developing new knowledge in metrology and assessing the occupational exposure and effectiveness of

means of prevention. The dissemination of information via a wide range of strategies designed to reach all work environments is an essential way to implement safe practices in the workplace. Orientations 2, 7, 8, 9 and 10 described above are consistent with these priorities.

- Finally, continue its leadership role at the provincial, Canadian and international levels, particularly through the transfer and validation of research results and its involvement in various targeted work groups, including those aiming to develop an ISO international standard, an approach to control banding (AFSSET/AFNOR/ISO) and a CSA Canadian standard.

13. DISCUSSION

A synthesis of existing occupational health and safety knowledge is required

While the R & D designed for the development of new products, applications in industry and marketing has been torrid for over a decade, research aiming to understand the health effects of occupational exposure or the risks of nanoparticles to occupational safety is at a less advanced state. Given this context, the introduction of nanomaterials raises several questions regarding their health and safety risks, especially in the workplace. In 2006, we published a literature review reporting on the scientific knowledge published up to the end of 2004. This updated second edition covers the scientific literature published up to early 2010. Consequently, certain general sections have been abridged; readers seeking a more detailed version of these sections can consult the first edition. In this new edition, one notes, on the one hand, the still significant lack of scientific knowledge relating to the different essential aspects for risk evaluation and control. Nevertheless, the rapid increase in research specific to NP carried out in different laboratories over the last few years substantially enriches the first edition of the report.

New materials with unique properties, the fruit of a multidisciplinary approach to research

An industrial revolution in nanotechnology (NT) is underway. The enthusiasm for the potential applications of NT may be gauged by noting the remarkable investment in research in this field over the last few years. NT does not alone represent another step towards miniaturization. At this scale, certain nanoparticles reveal unique properties (resistance, hardness, adhesion…) that are radically different from substances with the same composition but with larger dimensions. In this context, our definition of NP specifically targeted voluntarily synthesized products from the standpoint of using the unique properties demonstrated at these dimensions. However, ultrafine particles of comparable dimensions resulting from different processes such as emissions from diesel motors or welding fumes were not retained.

Eventually, the overall impact of NT is likely to revolutionize every sphere of society: science, industry, the environment, human health… Indeed, we can expect a plethora of applications in every field of human activity from the biomedical field to electronics, by way of metallurgy, agriculture, textiles, coatings, cosmetics, energy, catalysts, etc. Anticorrosion materials, self-cleaning windows, drugs delivered directly to the affected site or anti-graffiti paints represent only a few examples of potential applications. More than 1000 products are already marketed and organizations estimate that the annual market for “nano” products will reach 1,000 billion US dollars by 2012-2013, generating considerable economic spinoffs while creating employment for approximately two million people.

Many Québec workers potentially exposed

Québec has about fifty research teams consisting of about 200 professor-scholars active in this field and over 1,000 students divided up amongst almost all the province’s universities, four CEGEPS and several research centres. The NanoQuébec website lists nearly fifty enterprises in Québec currently working in nanotechnology. In addition, we need to take into account that Québec imports nanoparticles for a variety of fields in which workers are already potentially exposed. It is expected that over the next few years there will be an increase in the number of Québec workers who, because they use, transform or produce NP, are exposed to them.

Furthermore, a few new NP production and NP integration plants were in the start-up phase at the time this document was being written. Even if it is relatively easy to identify researchers and industrial plants producing NP, the identification of industrial plants buying NP and incoporating them in their production lines is still a challenge, making it difficult to sustain the implementation of safe work practices in these enterprises.

Various health effects have already been documented

Despite the growing number of published studies, the corpus of current knowledge on the health risks and effects of engineered NP is still incomplete. We are therefore currently in an important paradigm situation. In fact, on the one hand, the use of new, often unexpected properties of NP offers an extraordinary potential for economic development with applications in many fields. On the other hand, our inability to anticipate the behaviour of NP in the human body, due to their new and often unforeseeable properties, is at the centre of our concerns. Of primary concern in the present document are NP that are insoluble or only slightly soluble in biological fluids, since the toxicity of soluble particles is related solely to their chemical composition and to their deposition sites. The toxicity of soluble products is usually well known and independent of the particle's original size.

For NP that are insoluble or only slightly soluble in biological fluids, some major trends seem to be emerging. Several studies have shown behavior that is unique to NP because of their tiny size. Some NP can pass through our various defence mechanisms and be transported into the organism in insoluble form. Translocation, the ability to move to other sites in the body, is an important characteristic of insoluble nanoparticles. Thus, they can be found in small proportions in the blood and lymph systems after passing through the pulmonary or gastrointestinal membranes, spread to the different organs and accumulate at certain specific sites. Other NP, captured in the nasal passages, can travel along olfactory nerves, pass through the hematoencephalic barrier and directly penetrate the brain, or cross the cell barriers and reach the nucleus of the cell. Some NP can breach the placental barrier and lodge in the embryo. Widely studied in pharmacology, these properties could allow use of NP as vectors to carry drugs to targeted sites, including the brain. The corollary is that undesirable NP could be distributed in the organism of exposed workers and produce deleterious effects. Our knowledge of the human body's ability to recognize these foreign bodies and take charge of them adequately is still limited.

Normally, in toxicology, the effects are correlated to the quantity of product to which animals or humans are exposed. The greater the absorbed mass, the greater the effect. In the case of NP, it has been shown clearly that the measured effects are not well correlated to the product's mass, thus upsetting the classical interpretation of toxicity measurements. It is averred that, at equal mass, NP normally are more toxic than larger products of the same chemical composition. A consensus seems to be emerging in the scientific community that several factors contribute to the toxicity of these new generation products and that it is currently impossible, based on our fragmentary knowledge, to weigh their relative importance or predict the precise toxicity of a new NP.

For a specific chemical composition, many published studies relate the observed effects to the specific surface, surface properties, number, size and granulometric distribution of the NP. Concentration, surface dose, coating and surface properties, degree of agglomeration of the particles and pulmonary deposition site, surface charge, shape, porosity, crystalline structure, electrostatic attraction potential, particle synthesis method, hydrophilic/hydrophobic character and

post-synthesis modifications are other factors that influence NP toxicity. The presence of certain contaminants, such as metals, can also favour formation of free radicals and inflammation, while chemical composition and release of surface components, NP colloidal and surface properties, compartmentation in the airway and biopersistence are all factors that make understanding their toxicity even more complex. Moreover, the biopersistence of certain NP or certain components of NP in the body is becoming a major factor in their toxicity. These various factors consequently influence their functional, toxicological and environmental impact.

Several effects have already been shown in animals, including toxic effects on different organs (heart, lungs, kidneys, reproductive system…), as well as genotoxicity and cytotoxicity. Since the main absorption route in the work environment is still the airway, different studies clearly show that the very small size of nanoparticles is a key factor in toxicity, especially for particles that are insoluble or only slightly soluble.

Some NP can induce growth of reactive oxygen species, cause oxidizing stress, generate activation and injury of the airway cells, or cause inflammation, allergy, infection, bronchial obstruction and tissue damage, all possibly leading to changes in respiratory function. Some particles, for example, cause granulomas, fibrosis and tumor reactions at the pulmonary level in rat studies. The data currently available also suggest that cutaneous absorption would be relatively limited. Although major trends are emerging and reveal numerous toxic effects related to certain NP, it is found that each synthesized product can have its own toxicity. It seems that any modification of a process or a surface can have an impact on the resulting product's toxicity. Unfortunately, a very large proportion of the toxicological data currently available on several NP is difficult to extrapolate under real occupational exposure conditions because the exposure methodologies used are very different from the workplace realities. For example, few pulmonary toxicity studies have been carried out following the inhalation of airborne NP.

A current major concern of the scientific community involves CNT, which are very biopersistent. In fact, several recent studies suggest that carbon nanotubes would have potential effects similar to asbestos and that the toxicity would increase with the length and the accumulated dose. However, a major question remains unanswered: Would workplace exposure levels promote the inhalation of substantial quantities of CNT or does the application of current exposure control measures cause the workers' exposure to remain always at very low levels, even without exposure?

Assessment of the risk for workers

The toxic risk is based on the inherent toxicity of each NP and the level of exposure to this substance. In general, the toxicology data specific to each NP are still limited. The recent development of NT and the lack of information on working conditions and occupational exposure also greatly limit our ability to prognosticate the potential impact of NP exposure on workers' health. The information currently available gives reason to conclude that the main production processes can lead to occupational exposure if appropriate control measures are not implemented. Moreover, to avoid aggregation of particles, several processes have a post-synthesis stage to modify the particle's surface, often by coating it with another organic or inorganic substance; this operation has an impact on the nanoparticle's toxicity.

The technologies normally used in industrial hygiene are poorly adapted to characterization of nanoparticle levels in the work environment and restrict our ability to measure occupational

exposure, whereas the toxicological studies show that toxicity depends on several characteristics of nanoparticles — including surface, size, number, chemical composition and shape — and not only on mass. The development of strategies and tools allowing measurement of these additional parameters then becomes essential to risk assessment.

It must also be considered that because of their large specific surface, some nanoparticles offer major reactivity potential, which can result in fires or explosions ou uncontroled catalytic reactions. Specific prevention measures must be put in place to avoid such events. In the context of the current uncertainty, the risk will be impossible to quantify. A control banding approach can help determine the level of control to be established but requires a detailed analysis of each workstation and a good level of expertise.

Moreover, several occupational health and safety and environmental research organizations, such as NIOSH in the United States, HSE in England, JNIOSH in Japan or INRS in France as well as the IRSST, are currently conducting research to develop new knowledge eventually allowing quantitative risk assessments of workers' exposure. For this purpose, tools allowing assessment of occupational exposure must be adapted to the measurement characteristics required, depending on the nanoparticles' toxicity. Real exposure levels must be measured in the work environments and the effectiveness of the means of control and personal protection equipment must be evaluated.

Prevention: an essential aspect to implement

In such a context, this report is intended primarily as an information tool for Québec preventionists. It particularly provides information on the means and tools allowing people to work safely with engineered nanoparticles to prevent the risks of accidents or development of occupational diseases. This information is sometimes specific to nanoparticles, but most often relates to ultrafine particles, based on which analysis and intervention are projected. The best practices guide recently published by the IRSST completes this document.

Control of occupational exposure to nanoparticles includes several unknown factors. Indeed, the existing approaches to control by enclosure or respiratory protection during inhalation should be effective, but different aspects of this effectiveness still have to be proved. Control of exposure by ventilation at the source or general ventilation poses a challenge, because nanoparticles tend to behave more like a gas than like a solid. The data currently available tend to demonstrate their effectiveness but the studies must continue, mainly regarding the effectiveness of respiratory protection under actual conditions of use. Almost no data exist for personal protection against cutaneous exposure. Disposable Tyvek® clothing is recommended in a context in which it is likely that the methods based on most of the other existing personal protection equipment do not allow adequate protection.

Even in the absence of all of the knowledge required to assess the risk or the effectiveness of the available means of control, some countries are reconsidering their legislation and recommend a precautionary approach to protect workers from potential exposure and prevent the development of occupational diseases. Canada has also begun to legislate in the NP field. Canadian federal law obliges companies and institutions that produced or imported more than one kilogram of some specific nanomaterials in 2008 to submit all the information they have on these products: physical and chemical properties, toxicological data, manufacturing methods and uses.

Until their effects on the environment or on humans are documented adequately, strict prevention measures should be taken as a precaution to limit emission of these products both inside and outside the work environment.

Recommended follow-up

Finally, the research needs are discussed and avenues for prioritizing investment in research are proposed to the IRSST, in relation to its mandate, which is precisely to contribute to prevention by scientific research. In such a context, the authors of this report consider that the IRSST should favour the introduction of strict prevention procedures, which is still the only way to prevent the risk of development of occupational diseases or the occurrence of accidents. They thus strongly recommend that the IRSST concentrate its priority research efforts on the development of exposure assessment strategies and tools and the development and measurement of the effectiveness of means of controlling occupational NP exposure. Monitoring the scientific literature would allow advances in knowledge of NP toxicity, determination of the best parameters to measure for risk assessment and new developments in industrial hygiene to be taken into account. Efforts in knowledge transfer towards the various clients must continue, and even increase.

14. CONCLUSION

NT and NP constitute a fast-expanding field of activity; this is primarily due to the fact that NP have chemical and physical properties radically different from substances with the same composition but of a larger size. It is for this very reason that current technological development in this field is trying to turn these new and often unique properties to its advantage. At the same time, this constitutes a major challenge in OSH. Indeed, how does one assess the risks associated with these new products, and prevent the rise of occupational disease or new safety problems when the information available is extremely limited and totally insufficient?

The colossal sums invested in R & D in all industrialized countries will inevitably translate into discoveries of major importance whose applications will, in the near future, irreversibly affect our daily lives. Although their impacts are not yet known, more than 1000 products have already been commercialized, (cosmetics, automobile paints, waterproofers, textiles, etc.). The number and variety of workers at risk will increase in the coming years. If preventive measures in OSH are applied now to the field of NT, there will be significant benefits, since prevention and monitoring can be carried out at the design and implementation stages of a number of processes. However, given the fragmentary state of current knowledge regarding its OSH risks, this remains a major challenge.

Some NT applications will pose few new risks to the health and safety of workers. An example is electronics, where there have been constant advances in miniaturization, now at the nanometric level; in this environment, the risks are adequately controlled. On the other hand, there are real concerns about airborne free NP associated with other processes. These can cause OSH problems, or result in an accumulation of NP in the environment or enrichment through the food chain; this could lead to long-term risks to the health of populations. In spite of the currently limited knowledge on NP toxicity and the potential levels of worker exposure, the preliminary results obtained from most studies seem to indicate significant biological activity and undesirable deleterious effects for several NP. The proliferation of new NP and the modifications made to their surfaces, which will have a major impact on their surface properties and potentially their biological reactivity and toxicity, will make it virtually impossible in the short term to accumulate sufficient knowledge on the risks associated with each of these new particles.

In terms of occupational health, initiatives should be taken as soon as possible, using the best measurement tools and despite their limitations, to estimate levels of occupational exposure. Given the impossibility of developing quantitative risk assessments for each of these particles, it will be important to develop a precautionary approach, introduce prevention strategies, foster good work practices and avoid occupational risks so that we may prevent an increase in occupational disease and accidents. In addition, given the rapid evolution of knowledge in the field of OSH, it is essential to update IRSST reports in the near future.

15. BIBLIOGRAPHY

AFSSET, 2008. Les nanomatériaux. Avis de l'AFSSET Sécurité au travail. Agence française de sécurité sanitaire de l'environnement et du travail, July 2008, Paris, 247 p.

AFSSET, 2006. Les nanomatériaux : effets sur la santé de l'homme et sur l'environnement, Agence française de sécurité sanitaire de l'environnement et du travail, July 2006, Paris, 248 p.

Agence Nationale de la Recherche, 2005. « Santé-environnement et santé-travail : nouvelles perspectives de recherches », Séminaire de prospective scientifique et de lancement du programme recherche du Plan national santé environnement et du Plan santé travail, Ministère délégué à la recherche, République Française, 31 March et 1st April 2005, 37 p.

AIHA, 2007. Guidance for conducting Control Banding analysis, AIHA Guideline 9, American Industrial Hygiene Association, Fairfax, Virginia, 95 p.

Aitken RJ, Hankin SM, Ross B, Tran CL, Stone V, Fernandes TF, Donaldson K, Duffin R, Chaudhry Q, Wilkins TA, Wilkins SA, Levy LS, Rocks SA, Maynard A, 2009. EMERGNANO: A review of completed and near completed environment, health and safety research on nanomaterials and nanotechnology, publié par l'IOM, March 2009, 81 p.

Aitken RJ, Creely KS, Tran CL, 2004. Nanoparticles: An Occupational Hygiene Review, Institute of Occupational Medicine, Health and Safety Executive (HSE), UK, Research Report 274, 113 p.

Akerman ME, Chan WCW, Laakkonen P, Bhatia SN, Ruoslasti K, 2002. Nanocrystal targeting in vivo. *Proceedings of the National Academy of Sciences,* 99 (20) : 12617-21.

ASTM, 2006. Standard terminology Relating to Nanotechnology, Norme ASTM E2456-06, 4 p.

Balazy A, Toivola M, Reponen T, Podgórski A, Zimmer A and Grinshpun SA, 2006. Manikin-Based Performance Evaluation of N95 Filtering-Facepiece Respirators Challenged with Nanoparticules, *Annals of Occupational Hygiene Advance Access* published on December 12, 2005, *Annals of Occupational Hygiene* 2006, 50 : 259-269; doi : 10.1093/annhyg/mei058.

Balbus JM, Florini K, Denison RA, Waslsh SA, 2006. Getting it right the first time – Developping nanotechnology while protecting workers, public health and the environment, *Ann NY Acad Sci,* 1076 : 331-342.

Baron PA, Deye GJ, Chen BT, Schwegler-Berry DE, Shvedova AA, Castranova V, 2008. Aerosolization of Single-Walled Carbon Nanotubes for an Inhalation Study. *Inhalation Toxicology*, 20 (8) : 751- 760.

Bartis JT, Landree E, 2006. Nanomaterials in the Workplace. Policy Planning Workshop on Occupational Safety and Health, Report from Rand Corporation, 34 p.

BAUA, 2007. Guidance for Handling and Use of Nanomaterials at the Workplace. http://www.vci.de/default2~rub~809~tma~0~cmd~shd~docnr~121306~nd~~ond~nano~snd~~shmode~.htm.

Bégin D, Gérin M, 2002. *Substitution : démarche et outils*. *In* Solvants industriels – santé – sécurité – substitution, sous la direction de M. Gérin, Éd. Masson, Paris, p. 39-60.

Bello D, Brian L, Wardle, Yamamoto N, Guzman deVilloria R, Garcia EJ, Hart AJ, Ahn K, Ellenbecker MJ, Hallock M, 2009. Exposure to nanoscale particles and fibers during machining of hybrid advanced composites containing carbon nanotubes. *J Nanopart Res* 11 : 231–249.

Bémer D, Régnier R, Callé S, Thomas D, Simon X, Appert-Collin JC, 2006. Filtration des aérosols – Performances des médias filtrants, *Hygiène et Sécurité du Travail n°202*, cahier de notes documentaires, INRS, March 2006, p. 7-19.

Berges M, Mohlmann C, Welter J, Bard D, Mark D, Brouwer D, Stuurman B, Nieboer-op de Weegh M, Jankowska E, 2009. Exposure estimation from aerosol measurements in nanotechnology industries, 2009. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Bermudez E, Mangum JB, Wong BA, Asgharian B, Hext PM, Warheit DB, Everitt JI, 2004. Pulmonary Responses of Mice, Rats, and Hamsters to Subchronic Inhalation of Ultrafine Titanium Dioxide Particles. *Toxicological Sciences*, 77 : 347-357.

Birch ME, Evans DE, Ku BK, Ruda-Eberenz T, 2009. Air contaminants in a carbon nanofiber manufacturing facility. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Biswas P, Wu CY, 2005. Nanoparticules and the Environment, *Journal of the Air & Waste Management Association*, 55 : 708-746.

Borm PJ, Höhr D, Steinfartz Y, Zeitträger I, Albrecht C, 2000. Chronic inflammation and tumor formation in rats after intratracheal instillation of high doses of coal dusts, titanium dioxides, and quartz. *Inhalation Toxicology*, 12 (Supp. 3) : 225-231.

Bouillard J, Vignes A, Duffaud O, Perrin L, Thomas D, 2008. Explosion Risks from Nanomaterials. NanoSafe Conference, 6 November 2008, Grenoble.

Brenneman KA, Wong BA, Buccellato MA, Costa ER, Gross EA, Dorman DG, 2000. Direct olfactory transport of inhaled manganese ($^{54}MnCl_2$) to the rat brain: toxicokinetic investigations in a unilateral nasal occlusion model. *Toxicology and Applied Pharmacology* 169 : 238-248.

British Standards Institute (BSI), 2008. Nanotechnologies – Part 2: Guide to safe handling and disposal of manufactured nanomaterials. PD 6699-2 : 2007, BSI, London.

Brouwer DH, Gijsbers JHJ, Lurvink MWM, 2004. Personal Exposure to Ultrafine Particles in the Workplace: Exploring Sampling Techniques and Strategies, *Annals of Occupational Hygiene, 48 (5) : 439-453.*

Brouwer DH, Boeniger MF, Van Hemmen J, 2000. Hand wash and manual skin wipes, *Annals of Occupational Hygiene* 44 (7) : 501-510.

Brown DM, Donaldson K, Stone V, 2002a. Role of Calcium in the Induction of TNFα Expression by Macrophages on Exposure to Ultrafine Particles. *The Annals of Occupational Hygiene*, 46 (suppl 1) : 219-222.

Brown JS, Zeman KL, and Bennett WD, 2002b. Ultrafine Particle Deposition and Clearance in the Healthy and Obstructed Lung. *American Journal of Respiratory and Critical Care Medicine* 166 : 1240-1247.

Brown DM, Wilson MR, MacNee W, Stone V, Donaldson K, 2001. Size-Dependent Proinflammatory Effects of Ultrafine Polystyrene Particles: A Role for Surface Area and

Oxidative Stress in the Enhanced Activity of Ultrafines. *Toxicology and Applied Pharmacology*, 175 : 191-199

Brunner TJ, Wick P, Manser P, Spohn P, Grass RN, Limbach LK, Bruinink A, Stark WJ, 2006. In vitro cytotoxicity of oxide nanoparticles: comparison of asbestos, silica, and the effect of particle solubility. *Environmental Science & Technology* 40, 4374-4381.

Bruschi S, Thomas S, 2006 A Review of the Potential Occupational Health and Safety Implications of Nanotechnology. Flinders Consulting Pty Ltd for the Australian Safety and Compensation Council (ASCC), July 2006.

Bruske-Hohlfeld I, Peters A, Wichmann HE, 2005. Epidemiology of nanoparticles. Proceedins of the First International Symposium on Occupational Health Implications of Nanomaterials, October 12-14 2004, Buxton, UK, Health and Safety Executive Eds. and the National Institute for Occupational Safety and Health, United-States, July 2005, p 53-58.

Brouwer D, Stuurman B, Welter J, Moehlmann C, Berges M, Bard D, Wake D, Jankowksab E, Mark D, 2009. The NANOSH database for results of workplace air monitoring studies to manufactured nanomaterials (MNM): a preliminary tool to estimate the potential for inhalation exposure. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

BSI, 2005. Vocabulary nanoparticles, British Standards, PAS 71: 2005, 26 p.

Bureau of National Affairs, Washington D.C., 2004. Nanotechnology. *Conference Report*, Vol. 34, no. 42, p. 1068-1071.

Busnaina A, 2007. Nanomanufacturing Handbook, CRC Press, 400 p.

Castranova V, 2009. Pulmonary response to multi-walled carbon nanotube exposure, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

CEN, 2005. Workplace exposure–measurement of dermal exposure–principles and methods, TC137/WG 6 (prCEN/TR 15279). Berlin, Germany: European Committee for Standardization.

Chalupa DC, Morrow PE, Oberdörster G, Utell MJ, Frampton MW, 2004. Ultrafine particle deposition in subjects with asthma. *Env Health Perspec* 112 (8) : 879-882.

Chen CW, Huang SH, Chang CP, Chen CC, 2006a. Penetration of 4.5 nm to 10 μm Aerosol Particles through Fibrous Filters, Poster AIHCE, 2006.

Chen CW, Huang SH, Chang CP, Chen CC, 2006b. Penetration of 4,5 nm to 10 μm Aerosol Particles Through Fibrous Filters, Institute of Occupational Safety & Health, AIHA's Aerosol Technology Committee, Nanotechnology Symposium: Nanoparticles in the Workplace, May 13, 2006, Chicago, 4 p.

Chou YL, Ho CE, Tsai CJ, Chen CW, Chang CP, Shih TS, Chien CL, 2009. Characteristics of particles emitted from nanopowders dispersed using different methods, 2009. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Chow JC, Watson JG, Review of measurement methods and compositions for ultrafine particles, 2007. *Aerosol and Air Quality Research,* 7 (2) : 121-173.

CNRC, 2009. Nanotechnology, http://www.nrc-cnrc.gc.ca/randd/areas/nanotechnology_e.html.

Colvin VL, 2003a. Nanotechnology Research and Development Act of 2003, testimony before the U.S. House of Representatives Committee on Science, April 9, 2003. http://www.house.gov/science/hearings/full03/apr09/colvin.htm.

Colvin VL, 2003b. The Potential Environmental Impact of Engineered Nanomaterials, *Nature Biotechnology*, 21(10) : 1166-1170.

Commission de l'éthique de la science et de la technologie, 2006. Éthique et nanotechnologies : se donner les moyens d'agir. Commission de l'éthique de la science et de la technologie, Gouvernement du Québec, 152 p.

Commission européenne, 2007. European Commission is world's largest public investor in nanotechnology. Press release issued 13 September 2007. http://cordis.europa.eu/nanotechnology/.

Commission européenne, 2002. *Le 6e Programme-cadre de la Commission européenne, document synthèse publié par la Commission européenne.* Luxembourg, Office of the official publications of the European Communities, 52 p. http://www.cordis.lu/fp6/nmp.htm.

Commission des communautés européennes, 2005. Nanosciences et nanotechnologies: un plan d'action pour l'Europe 2005-2009. Published by the Commission of the European Communities, Brussels, 7 June 2005, 12 p. ftp://ftp.cordis.lu/pub/nanotechnology/docs/nano_action_plan2005_fr.pdf.

Conseil des académies canadiennes, 2008. Petit et différent : perspective scientifique sur les défis réglementaires du monde nanométrique. July 2008, 151 p.

Council for Science and Technology, 2007. Nanotechnologies review. http://www2.cst.gov.uk/cst/business/nanoreview.shtml.

CSA, 2010. Nanotechnologies – Health and safety practices in occupational settings. Temporary title, working document, Canadian Standards Association.

CSN, 2009. Le 28 avril, journée de commémoration. Perspectives CSN, March 2009, p 27.

Daigle CC, Chalupa DC, Gibb FR, Morrow PE, Oberdörster G, Utell MJ, 2003. Ultrafine particle deposition in humans during rest and exercise. *Inhalation Toxicology*, 15 : 539-552.

DEFRA, 2007. Characterising the Potential Risks posed by Engineered Nanoparticles. A Second UK Government Research Report Department for Environment, Food and Rural Affairs, 100 p.

De Guire L, Camus M, Case B, Langlois A, Laplante O, Lebel G, Lévesque B, Rioux M, Siemiatycki J, 2003. Épidémiologie des maladies reliées à l'exposition à l'amiante : rapport. Sous-comité sur l'épidémiologie des maladies reliées à l'exposition à l'amiante. INSPQ. INSPQ-2003-046.

Demou E, Peter P, Hellweg S, 2008. Exposure to Manufactured Nanostructured Particles in an Industrial Pilot Plant. *Ann Occup Hyg*, 52 (8) : 695–706.

Department for Environment, Food and Rural Affairs (DEFRA), 2005. Characterising the potential risks posed by engineered nanoparticles, A first UK Government research report, 60 p. http://www.defra.gov.uk/environment/nanotech/nrcg/pdf/nanoparticles-riskreport.pdf.

Dhaniyala S, Liu BYH, 1999. Investigations of Particle Penetration in Fibrous Filters. *Journal of the Institute of Environmental Sciences and Technology,* 42 (1) : 32-40.

Dinyer J, Turnbull M, Neale S, 2005. Risk Management of the Explosive Dusts in the Pharmaceutical Industry: A Practical Approach, *Pharmaceutical Technology,* August 1, 2005.

Dockery DW, Pope CA, Xu X, Spengler JD, Ware JH, Fay ME, Ferris BG, Speizer FE, 1993. An association between air pollution and mortality in six U.S. cities. *New England Journal of Medicine,* 329 (24) : 1753-1759.

Donaldson K, Murphy F, Poland C, 2009. High aspect ratio nanoparticles: the hazard from long biopersistent fibres, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Donaldson K, 2005a. The toxicology of airborne nanoparticules. Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials, october 12-14 2004, Buxton, UK, Health and Safety Executive eds. and the National Institute for Occupational Safety and Health, United States, July 2005, p 30-34.

Donaldson K, Tran L, Jimenez LA, Duffin R, Newby DE, Mills N, MacNee W, Stone V, 2005b. Combustion-derived Nanoparticles: a Review of Their Toxicology Following Inhalation Exposure, *Particle and Fibre Toxicology*, 2 : 10.

Donaldson K, Stone V, Gilmore PS, Brown DM, MacNee W, 2000. Ultrafine particles: mechanisms of lung injury. *Philosophical Transactions of The Royal Society of London, Series A 358*, p 2741-2749.

Donaldson K, Li XY, MacNee W, 1998. Ultrafine (nanometer) particle mediated lung injury. *Journal of Aérosol Science,* 29 (5-6) : 553-560.

Dorman DC, Brenneman KA, McElveen AM, Lynch SE, Roberts KC, Wong BA, 2002. Olfactory transport: a direct route of delivery of inhaled manganese phosphate to the rat brain. *Journal of Toxicology and Environmental Health,* 65 : 1493-1511.

Driscoll KE, 1996. Role of inflammation in the development of rat lung tumors in response to chronic particle exposure. *Inhalation Toxicology,* 8 (Suppl): 139-153.

Duffin R, Tran CL, Clouter A, Brown DM, MacNee W, Stone V, Donaldson K, 2002. The Importance of Surface Area and Specific Reactivity in the Acute Pulmonary Inflammatory Response to Particles, *The Annals of Occupational Hygiene*, 46 (Suppl 1) : 242-245.

Dupuy JP, Roure F, 2004. Les nanotechnologies: éthique et prospective industrielle, tome I, Conseil général des Mines, Paris, 73 p.

Edwards R, Smith KR, Kirby B, Alles T, Litton CD, Hering S, 2008. An inexpensive dual-chamber particle monitor: laboratory characterization. *J. Air & Waste Manage Assoc,* 56 : 789-799.

Eklund P, Ajayan P, Blackmon R, Hart AJ, Kong J, Pradhan B, Rao A, Rinzler A, 2007. International Assessment of Research and Development of Carbon Nanotube Manufacturing and Applications, *World Technology Evaluation Center, Inc.*, 4800 Roland Avenue, Baltimore, Maryland 21210, juin 2007, 138 pages.

Elder ACP, Gelein R, Azadniv M, Frampton M, Finkelstein J, Oberdörster G, 2004. Systemic effects of inhaled ultrafine particles in two compromised, aged rat strains. *Inhalation Toxicology*, 16 : 461-471.

Elder ACP, Gelein R, Azadniv M, Frampton M, Finkelstein J, Oberdörster G, 2002. Systemic interactions between inhaled ultrafine particles and endotoxin. *Annals of Occupational Hygiene,* 46 (Suppl 1) : 231-234.

Elder ACP, Gelein R, Finkelstein JN, Cox C, Oberdörster G, 2000. Pulmonary inflammatory response to inhaled ultrafine particles is modified by age, ozone exposure, and bacterial toxin. *Inhalation Toxicology*, 12 (Suppl. 4) : 227-246.

Ellenbecker M, 2007. Interim best practices for working with nanoparticles. Center for high-rate nanomanufacturing, ASTM E2456-06 Draft 3, 101-2-2007.

Endo M, Hayashi T, Itoh I, Kim YA, Shimamoto D, Muramatsu H, Shimizu Y, Mansfeld F, Jeanjacquet SL, 1986. An Anticorrosive Magnesium/Carbon Nanotube Composite. Corrosion Science, 26 : 727.

Eninger RM, Honda T, Reponen T, McKay R, Grinshpun SA, 2008a. What does respirator certification tell us about filtration of ultrafine particles? *J Occup Environ Hyg*, *5* : 286–295.

Eninger RM, Honda T, Adhikari A, Heinonen-Tanski H, Reponen T, Grinshpun SA, 2008b. Filter performance of N99 and N95 facepiece respirators against viruses and ultrafine particles. *Ann Occup Hyg,* 52 (5) : 385–396.

Environmental Defense – DuPont Nano Partnership, 2007. NANO Risk Framework, June 2007, 104 p.

EPAQS, 2001. Airborne Particles. Expert Panel on Air Quality Standards. The Stationery Office Limited, UK. ISBN 0 11 753599 0.

European Nanotechnology Gateway, 2004. EU and nanotechnology in the new member states and the candidate countries, Who`s who and Research Priorities, Nanoforum consortium, 13 p.

Evans DE, Ku BK, Birch ME, Dunn KH, 2009. Direct reading monitoring of contaminants in a carbon nanofiber manufacturing facility. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Faunce TA, 2007. Nanotherapeutics: new challenges for safety and cost-effectiveness regulation in Australia, *MJA*, 186 (4) : 189-191.

Faux SP, Tran CL, Miller BG, Jones AD, Monteiller C, Donaldson K, 2003. In Vitro Determinants of Particulate Toxicity : The Dose-Metric for Poorly Soluble Dusts, Institute of Occupational Medicine for the Health and Safety Executive 2003, Research Report 154, 64 p.

FDA, 2007. Nanotechnology A Report of the U.S. Food and Drug Administration Nanotechnology Task Force, US FDA, July 2007.

Ferin J., Oberdörster G., Penney DP, 1992. Pulmonary retention of ultrafine and fine particles in rats. *American Journal of Respiratory Cell and Molecular Biology,* 6 : 535-542.

Finnish Institute of Occupational Health, 2007. Nanosh Inflammatory and Genotoxic Effects of Engineered Nanoparticles. NanoSafety Hub Meeting, FIOH, 23 March 2007.

Fissan H, Neumann S, Trampe A, Pui DYH, Shin WG, 2007. Rationale and principle of an instrument measuring lung deposited nanoparticles surface area. *Journal Nanoparticle Research*, 9 : 53-59.

Frampton MW, Stewart JC, Oberdörster G, Morrow PE, Chalupa D, Pietropaoli AP, Frasier LM, Speers DM, Cox C, Huang LS, Utelle MJ, 2006. Inhalation of Ultrafine Particles Alters Blood Leukocyte Expression of Adhesion Molecules in Humans. *Environmental Health Perspectives*, 114 (1) : 51-58.

Fujitani Y, Kobayashi T, Arashidani K, Kunugita N, Suemura K, 2008. Measurement of the Physical Properties of Aerosols in a Fullerene Factory for Inhalation Exposure Assessment. *J Occ Env Hyg*, 5 : 380–389.

Gallagher J, Sams R, Inmon J, Gelein R, Elder A, Oberdörster G, Prahalad AK, 2003. Formation of 8-oxo-7,8-dihydro-2'-deoxyguanosine in rat lung DNA following subchronic inhalation of carbon black. *Toxicology and Applied Pharmacology*, 190 : 224-231.

Garshick E, Laden F, Hart JE, Smith TJ, Rosner B, 2006. Smoking imputation and lung cancer in railroad workers exposed to diesel exhaust. *Am J Ind Med*, 49 (9) : 709–718.

Garshick E, Laden F, Hart JE, Rosner B, Smith TJ, Dockery DW, Speizer FE, 2004. Lung cancer in railroad workers exposed to diesel exhaust. *Environ Health Perspect*, 112(15) : 1539–1543.

Gazette du Canada, 2009. *Gazette du Canada, Partie II*, Ottawa, 143 (5) : 257-259, 4 March 2009.

Geraci C, Mathner M, Hodson L, 2009. Evaluating nanoparticles emissions in the workplace : a description of the approach used by NIOSH and a summary of findings from 12 site visits. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Geraci, 2009. Nanotechnology : the new workplace – NIOSH research to meet the challenge, Congrès INNO09, Montréal, 24 April 2009.

Gérin M, Bégin D, 2004. *Substitution*. *In* Manuel d'hygiène du travail : Du diagnostic à la maîtrise des facteurs de risque, Éd. Modulo-Griffon, Montréal, p. 553-566.

Gilmour PS, Ziesenis A, Morrison ER, Vickers MA, Drost EM, Ford I, Karg E, Mossa C, Schroeppel A, Ferron GA, Heyder J, Greaves M, MacNee W, Donaldson K, 2004. Pulmonary and systemic effects of short-term inhalation exposure to ultrafine carbon black particles. *Toxicology and Applied Pharmacology*, 195 : 35-44.

Granier JJ, Pantoya ML, 2004. Laser ignition of nanocomposite thermites. *Combust Flame*, 138 : 373–382.

Gray CA, Muranko H, 2006. Studies of robustness of industrial aciniform aggregates and agglomerates-carbon black and amorphous silicas: A review amplified by new data. *Journal of Occupational Environmental Medicine*, 48 : 1279-1291.

Green TR, Fisher J, Stone MH, Wroblewski BM, Ingham E, 1998. Polyethylene particles of a 'critical size' are necessary for the induction of cytokins by macrophages in vitro. *Biomaterials*, 19 : 2297-2302.

Grinshpun SA, Haruta H, Eninger RM, Reponen T, McKay RT, Lee SA, 2009. Performance of an N95 filtering facepiece particulate respirator and a surgical mask during human breathing: two pathways for particle penetration, *J Occ Env Hyg*, 6 : 593-603.

Grinshpun SA, 2006. Nanoparticles: Aerosol measurement and respiratory protection, AIHA's Aerosol Technology Committee, *Nanotechnology Symposium: Nanoparticles in the Workplace*, May 13, 2006, Chicago, p. 57-81.

Han JH, Lee EJ, Lee JH, So KP, Lee YH, Bae GN, Lee S-B, Ji JH, Cho MH, Yu IJ, 2008. Monitoring multiwalled carbon nanotube exposure in carbon nanotube research facility. *Inhal Toxicol*, 20 (8) : 741–749.

Hanai S, Kobayashi N, Ema M, Ogura I, Gamo M, Nakanishi J, 2009. Risk assessment of manufactured nanomaterials : titanium dioxide (TiO_2), National Institute of Advanced Industrial Science and Technology (AIST), 56 p.

Hankin M, Tran CL, Ross B, Donaldson K, Stone V, Chaudhry Q, 2008. Cell Pen: A study to identify the physico-chemical factors controlling the capacity of nanoparticles to penetrate cells. Rapport IOM, projet CB0407, 12 August 2008, 43 p.

Hansen SF, 2009. Regulation and Risk Assessment of Nanomaterials – Too Little, Too Late? Thèse de doctorat, Université technique du Danemark, 130 p.

Harford AJ, Edwards JW, Priestly BG et Wright PFA, 2007. Current OHS best practices for the Australian Nanotechnology industry. NanoSafe Australia OHS Position Paper, November 2007, 18 p.

Hart JE, Laden F, Schenker MB, Garshick E, 2006. Chronic obstructive pulmonary disease mortality in diesel-exposed railroad workers. *Environ Health Perspect*, 114 (7) : 1013–1017.

Health and Safety Executive (HSE), 2007a. HSE NanoAlert Service, Issue 2. Health and Safety Executive, May 2007.

Health and Safety Executive (HSE), 2007b. NanoAlert Service, Issue 3, Health and Safety Executive, August 2007.

Health and Safety Executive (HSE), 2006. NanoAlert Service, Issue 1, Health and Safety Executive, Décember 2006.

Health and Safety Executive (HSE), 2004a. A review of the toxicity of particles that are intentionally produced for use in nanotechnology applications, seen from an occupational health perspective. HSE, 40 p.

Health and Safety Executive (HSE), 2004b. Health effects of particles produced for nanotechnologies. HSE Hazard assessment document EH75/6, UK, Décember 2004, 37 p.

Health and Safety Executive (HSE), 2004c. Nanotechnology, HSE information note no. HSIN1. Sudbury, Suffolk, G.-B. HSE, 2004, 4 p., http://www.hse.gov.uk/pubns/hsin1.pdf.

Heim M, Mullins B, Wild M, Meyer J, Kasper G, 2005. Filtration efficiency of aerosol particles below 20 nanometers. *Aerosol Sci Tech*, 39 : 782–789.

Heinrich U, Fuhst R, Rittinghausen S, Creutzenberg O, Bellmann B, Koch W, Levsen K, 1995. Chronic inhalation exposure of wistar rats and two different strains of mice to diesel engine exhaust, carbon black and titanium dioxide. *Inhalation Toxicology*, 7 : 533-556.

Helland A, Wick P, Koehler A, Schmid K, Som C, 2007. Reviewing the Environmental and Human Health Knowledge Base of Carbon Nanotubes. *Environmental Health Perspectives,* 115 (8) : 1125-1131.

Herber RFM, Duffus JH, Christensen JM, Olsen E, Park MV, 2001. Risk Assessment for Occupational Exposure to Chemicals. A Review of Current Methodology. *Pure and Applied Chemistry*, 73 (6) : 993-1031.

Hett A, 2004a. Nanotechnology: Small matters, many unknowns, Swiss Reinsurance Company, 57 p. http://www.swissre.com/INTERNET/pwsfilpr.nsf/vwFilebyIDKEYLu/ULUR-5YNGET/$FILE/Publ04_Nanotech_en.pdf.

Hett A, 2004b. Swiss Re on risk : nanotechnology. Armonk, N.Y. Corporate Communications. http://www.swissre.com/INTERNET/pwsfilpr.nsf/vwFilebyIDKE YLu/FSTN-5Z4MTD/%24FILE/Nanotechnology_US_brochure.pdf.

Hinds WC, 1999. Aerosol technology: properties, behavior, and measurement of airborne particles. 2nd Ed. New York: Wiley-Interscience.

Hoang KT, 1992. Dermal Exposure Assessment: Principles and Applications. U.S. Environmental Protection Agency, Office of Health and Environmental Assessment, Washington, DC, EPA/600/8-91/011B.

Höck J, Hofmann H, Krug H, Lorenz C, Limbach L, Nowack B, Riediker M, Schirmer K, Som C, Stark W, Studer C, von Götz N, Wengert S, Wick P, 2008. Guidelines on the Precautionary Matrix for Synthetic Nanomaterials. Federal Office for Public Health and Federal Office for the Environment, Confédération Switzerland, Berne, 28 p.

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

Hsu LY, Chein HM, 2007. Evaluation of nanoparticle emission for TiO_2 nanopowder coating materials. *Journal of Nanoparticle Research*, 9 : 157-163.

Huang CY, Tsai CJ, Huang CH, Chen CW, Chang CP, Shih TS, 2009a. Exposure study of respirable and nanosized particles in a pigment grade TiO_2 powder plant. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Huang CH, Tay CY, Huang CY, Tsai CJ, Chen CW, Shih TS, 2009b. Respirable and nanoparticle sampling and analysis in the workplace. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Hurt RH, Monthioux M, Kane A, 2006. Toxicology of carbon nanomaterials: status, trends, and perspectives on the special issue. *Carbon*, 44 : 1028-1033.

Ibald-Mulli A, Wichmann HE, Kreyling W, Peters A, 2002. Epidemiological evidence on health effects of ultrafine particles. *Journal of Aerospace Medicine*, 15 : 189-201.

Ichihara G, Li W, Fujitani Y, Ichihara S, Ding X, Liu Y, Wang Q, Sai U, Hata N, Kobayashi T, 2009. Exposure assessment and evaluation of health status in workers handling titanium dioxide, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

ICON 2008. Towards Predicting Nano-Biointeractions: An International Assessment of Nanotechnology Environment, Health and Safety Research Needs. *International Council on Nanotechnology, Number 4, May 1, 2008, 80 p.*

ICON, 2006. A Review of Current Practices in the Nanotechnology Industry – Phase two report: Survey of current practices in the nanotechnology workplace. University of California, Santa Barbara for the *International Council on Nanotechnology* (ICON), November 13, 2006.

Ignacio J. S., Bullock W. H., eds. 2006. A strategy for assessing and managing occupational exposures. 3rd Edn. Fairfax, VA: American Industrial Hygiene Association. ISBN 0 932627 86 2.

Illing P, 2006. General overview of the safety evaluation of chemicals. *Environmental Science and Technology,* 23 : 1-27.

Institute of Occupational Medecine (IOM), 2009. Review of occupational hygiene reports on suitability of respiratory protective equipment (RPE). Prepared for the Health and Safety Executive, Research Report 746, 46 p.

INRS, 2005. Évaluation du risque incendie dans l'entreprise/Guide méthodologique, ED 970, Paris, 30 p.

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

IRSST, 2009. Plan triennial de recherche – production scientifique et technique 2009-2011. IRSST, 2008 (internal document).

IRSST, 2006. Osons le changement, plan stratégique 2006-2010. IRSST, Montréal, 23 March 2006, 39 p (internal document).

ISO, 2009a. Guidance on what physico-chemical characterization of manufactured nanomaterials are used for toxicological assessment. Draft outline, Version October 5, 2009.

ISO, 2009. Nanotechnologies – Characterization of nanoparticles in inhalation exposure chambers for inhalation toxicity testing, Draft International Standard ISO/DIS10808.

ISO, 2008a. ISO/TS 27687 : 2008.

ISO, 2008b. Nanotechnologies – Health and safety practices in occupational settings relevant to nanotechnologies, ISO /TR 12885, rapport technique, October 2008, 79 p.

ISO, 2007. Workplace Atmospheres – Ultrafine, nanoparticle and nano-structured aerosols – Inhalation exposure characterisation and assessment, *ISO/TR* 27628 : 2007.

Jaques PA, Kim CS, 2000. Measurement of total lung deposition of inhaled ultrafine particles in healthy men and women. *Inhalation Toxicology,* 12 : 715-731.

Japuntich DA, Franklin LM, Pui DY, Kuehn TH, Kin SC, 2007. A comparison of two nano-sized particle air filtration tests in the diameter range of 10 to 400 nanometers. *Journal of Nanoparticle Research,* 9 : 93-107.

Jarvela M, Koivisto J, Lyyranen J, Lahtimaki M, Saamanen A, Heikkila K, Hameri K, Auvinen A, Jokiniemi J, Tuomi T, 2009. Exposure to ultrafine particles and nanoparticles in TiO_2 production plant, 2009. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health,* 26-29 August 2009, Helsinki.

Kagan VE, Tyurina YY, Tyurin VA, Konduru NV, Potapovich AI, Osipov AN, Kisin ER, Schwegler-Berry D, Mercer R, Castranova V, Shvedova AA, 2006. Direct and Indirect Effects of Single Walled Carbon Nanotubes on RAW 264.7 Macrophages: Role of Iron, *Toxicol Lett,* 165 (1) : 88-100.

Kaluza S, Honnert B, Jankowska E, Pietrowski P, Rosell MG, Tanarro C, Tejedor J, Zugasti A, 2008. Workplace exposure to nanoparticles. European Risk Observatory Report, 2008. EU-OSHA – European Agency for Safety and Health at Work, 89 p.

Kandlikar M, Ramachandran G, Maynard A, Murdock B, 2007. Health risk assessment for nanoparticles : A case for using expert judgment. *Journal of Nanoparticle Research,* 9 : 137-156.

Katz LC, Burkhalter A, Dreyer WJ, 1984. Fluorescent latex microspheres as a retrograde neuronal marker for in vivo and in vitro studies of visual cortex. *Nature,* 310 : 498-500.

Kim Y, Kim H, Park J, Kwak BK, Bae E, Lee J, Choi K, Lee B-C, Yi J, 2009. Exposure assessment to suspension of silver nanoparticles in lab-scale. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health,* 26-29 August 2009, Helsinki.

Kim SE, Harrington MS, Pui DYH, 2007. Experimental study of nanoparticles penetration through commercial filter media. *J. Nanopart. Res*, 9 : 117-125.

Kim CS, Bao L, Okuyama K, Shimada S, Niinuma H, 2006. Filtration efficiency of a fibrous filter for nanoparticles. *J. Nanopart. Res*, 8 : 215-221.

Kim CS, Jaques PA, 2000. *Phil Trans Roy Soc London* A 358 : 2693-2705.

Kirby DC, 2005. Back to the basics in dust explosions. Conférence présentée à l'American Institute of Chemical Engineers, Spring National Meeting, 11 to 13 April 2005.

Knaapen AM, Borm PJ, Albrecht C, Schins RPF, 2004. Inhaled Particles and Lung Cancer. Part A: Mechanisms, *International Journal of Cancer*, 109: 799-809

Knebel S, Meili C, 2010. Conference Report : 5th Int. "NanoRegulation" Conference, 25 – 26 November 2009, Rapperswil (Switzerland), The Innovation Society Ltd, 49 p.

Knight DJ, 2006. Intelligent approaches to safety evaluation. *Issues in Environmental Science and Technology*, 23 : 74-94.

Kobayashi N, Ogura I, Gamo M, Kishimoto A, Nakanishi J, 2009. Risk assessment of manufactured nanomaterials : carbon nanotubes (CNTs), National Institute of Advanced Industrial Science and technology (AIST), 48 p.

Kohler M, Fritzsche W, 2004. Nanotechnology, An Introduction to nanostructuring techniques. Weinheim, Germany ,Wiley-VCH, 272 p.

Koivisto AJ, Korhonen R, Jarvela M, Aromaa M, Lehtinen S, Hameri K, Makela JM, Pasanen P, 2009. Nanoparticle emissions from liquid flame spray processes. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health,* 26-29 August 2009, Helsinki.

Kreyling WG, Semmler M, Erbe F, Mayer P, Takenaka S, Schulz H, Oberdörster G, Ziesenis A, 2002. Translocation of ultrafine insoluble iridium particles from lung epithelium to extrapulmonary organs is size dependent but very low. *Journal of Toxicology and Environmental Health,* 65 (20) : 1513-1530.

Kreyling WG, Scheuch G, 2000. Clearance of Particles deposited in the lungs, *In* Particle lungs interactions (Gehr P, Heyder J, Eds), New-York – Basel: Marcel Dekker Inc, 323-376.

Kroll A, Pillukat MH, Hahn D, Schnekenburget J, 2009. Current *in vitro* methods in nanoparticles risk assessment : limitations and challenges. *Eur J Pharma and Biopharm*, 72 : 370-377.

Kroto HW, Heath JR, O'Brian SC, 1985. C_{60}: Buckminsterfullerene. *Nature*, 318 : 162-163.

Kuempel ED, 2006a. Quantitative Risk Assessment Methods for Nanoparticles: Strategies and Data Needs, Ist International Conference on Nanotoxicology: Biomedical Aspects, February 1, 2006, Miami, Florida.

Kuempel ED, Tran CL, Castranova V, Bailer AJ., 2006b. Lung Dosimetry and Risk Assessment of Nanoparticles: Evaluating and Extending Current Models in Rats and Humans. *Inhalation Toxicology*, 18 : 717-724.

Kuhlbusch, TAJ, Fissan H, Asbach C, 2009. Nanoparticles and Exposure: Measurement Technologies and Strategies, in "Nanomaterials: Risks and Benefits", I. Linkov and J. Steevens (Eds.), ISBN: 978-1-4020-9490-3, Springer, Dordrecht, The Netherlands, 233-247.

Kuhlbusch, TAJ, Fissan H, Asbach C, 2008. Measurement and detection of nanoparticles in the environment, in Nanotechnology, Volume 2: Environmental Aspects, Ed. H. Krug, ISBN 978-3-527-31735-6, Wiley-VCH, Weinheim, p. 229-266.

Kuhlbusch, TAJ, Fissan H, 2006. Particle Characteristics in the Reactor and Pelletizing Areas of Carbon Black Production, *J. Occ. Env. Health* 3 (10) : 558-567.

Kuhlbusch TAJ, Neumann S, Fissan H, 2004. Number Size Distribution, Mass Concentration, and Particle Composition of Pm_1, $Pm_{2.5}$, and PM_{10} in Bag Filling Areas of Carbon Black Production. *Journal of Occupational and Environmental Hygiene*, 1 : 660-671.

Kuzma J, 2007. Moving forward responsibly: oversight for the nanotechnology-biology interface. *Journal of Nanoparticle Research*, 9 : 165-182.

Lam CW, James JT, McCluskey R, Arepalli S, Hunter RL, 2006. A Review of Carbon Nanotube Toxicity and Assessment of Potential Occupational and Environmental Health Risks. *Critical Reviews in Toxicology*, 36 (3) : 189-217.

Lam CW, James JT, McCluskey R, Hunter RL, 2004a. Pulmonary toxicity of single-wall nanotubes in mice 7 and 90 days after intratracheal instillation. *Toxicological Sciences*, 77 : 126-134.

Lamy P, 2005. Nanomatériaux: Risques pour la santé et l'environnement. Centre de recherche sur la matière divisée, Orléans, France, 29 p.

Lara J, McCabe J, Vennes M, 2010. Guide des appareils de protection respiratoire utilisés au Québec, IRSST, Document in scientific evaluation, 154 p.

Lara J, Vennes M, 2003. Guide pratique de protection respiratoire, 2nd Edition, CSST, Montréal.

Lavoie J, Cloutier Y, Lara J, Marchand G, 2007. Guide sur la protection respiratoire contre les bioaérosols – Recommandations sur le choix et l'utilisation. Études et recherches, Guide technique RG-497, Montréal, IRSST, 40 p.

Lebeau D, 2001. *Aperçu de la recherche sur les nanotechnologies.* Sainte-Foy, Conseil de la science et de la technologie, 55 p.

Levelt WJM, 2004. How big can small actually be? Some remarks on research at the nanometer scale and the potential consequences of Nanotechnology. Study Group on the consequences of Nanotechnology. Royal Netherlands Academy of Arts and Sciences. 41 p.

Li XY, Gilmour P, Donaldson K, MacNee W, 1996. Free radical activity and proinflammatory effects of particulate air pollution (PM10) *in vivo* and *in vitro. Thorax,* 51 : 1216-1222.

Lindberg HK, Falck GCM, Suhonen S, Vippola M, Vanhala E, Catalan J, Savolainen K, Norppa H, 2009. Genotoxicity of nanomaterials : DNA damage and micronuclei induced by carbon nanotubes and graphite nanofibres in human bronchial epithelial cells in vitro. *Toxicology Letters*, 186 : 166-173.

Lison D, Lardot C, Huaux F, Zanetti G, Fubini B, 1997. Influence of particle surface area on the toxicity of insoluble manganese dioxide dusts. *Archives of Toxicology*, 71 (12) : 725-729.

Litton CD, Smith KR, Edwards R, Allen T, 2004. Combined optical and ionization measurement techniques for inexpensive characterization of micrometer and submicrometer aerosols, *Aerosol Sci and Technol*, 38 : 1054-1062.

Lorrain JL, Raoul D, 2004. Nanosciences et progrès médical Paris, Le Sénat, 2004. http://www.senat.fr/rap/r03-293/r03-2930.html.

Lovric J, Bazzi HS, Cuie Y, Fortin GRA, Winnik M, Maysinger D, 2005. Differences in subcellular distribution and toxicity of green and red emitting CdTe quantum dots. *Journal of Molecular Medicine*, 83 : 377-385.

Lux Research, 2007. The Nanotech Report, 5th Edition, New York: Lux Research.

MacNee W, XY Li, Gilmour P, Donaldson K, 2000. Systemic effect of particulate air pollution. *Inhalation Toxicology*, 12 (Suppl 3) : 233-244.

Mantovani E, Porcari A, Meili C, Widmer M, 2009. Mapping study on regulation and governance of nanotechnologies. FramingNano Report, January 2009, 138 p.

Mamberger JH, Kvamme EF, 2008. The National Nanotechnology Initiative : Second Assessment and Recommendations of the National Nanotechnology Advisory Panel, Report of the President's Council of Advisors on Science and Technology, April 2008, 56 p.

Marburger JH, Connaughton JL, 2007. Policy Principles For Nanotechnology Environmental, Health, And Safety Oversight, Executive Office of the President and Council On Environmental Quality Executive Office Of The President, Office Of Science And Technology, November 8, 2007.
http://www.ostp.gov/html/nano%20ehs%20principles%20memo_ostp-ceq_fincollpdf.

Mark D, Bard D, Wake D, Berges M, Mohlman C, Welter J, Brouwer D, Stuurman B, Nieboer-op de Weegh M, Jankowska E, 2009. Outcome of EU project NANOSH to assess potential worker exposure to engineered nanoparticles. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Mark D, 2005a. Control of nanoparticles, Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials, October 12-14 2004, Buxton, UK, Health and Safety Executive and the National Institute for Occupational Safety and Health, United States, July 2005, p. 78-83.
http://www.hsl.gov.uk/capabilities/nanosymrep_fincollpdf.

Mark D, 2005b. Nanomaterials a risk to health at work?, Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials, October 12-14 2004, Buxton, UK, Health and Safety Executive and the National Institute for Occupational Safety and Health, United States, July 2005, p 150-156.

Martin SB, Moyer ES, 2000, Electrostatic respirator filter media: filter efficiency and most penetrating particle size effects. *Appl Occup Env Hyg*, 15 (8) : 609-617.

Massachusetts Institute of Technology (MIT) 2006. Potential Risks of Nanomaterials and How to Safely Handle Materials of Uncertain Toxicity. http://web.mit.edu/environment/ehs/topic/nanomatericollhtml.

Maynard AD, Aitken RJ, 2007. Assessing exposure to airborne nanomaterials: current abilities and future requirements. *Nanotoxicology*, 1 (1) : 26-41.

Maynard AD, 2006. Nanotechnology : assessing the risks. *Nanotoday*, 1 (2) : 22-33.

Maynard AD, Kuempel ED, 2005. Airborne nanostructured particles and occupational health. *Journal of Nanoparticle Research*, 7 : 587-614.

Maynard AD, 2004. Nanotechnology – a new occupational health challenge for a new generation? ICOH, *Newsletter*, 2 (3) : 4-6.

Maynard AD, Baron PA, Foley M, Shvedova AA, Kisin ER, Castranova V, 2004. Exposure to Carbon Nanotube Material: Aerosol Release During the Handling of Unrefined Single-Walled Carbon Nanotube Material, Part A, *Journal of Toxicology and Environmental Health*, 67 (1) : 87-107.

Ménard L, 2004. Principes généraux de maîtrise. *In* Manuel d'hygiène du travail. Du diagnostic à la maîtrise des facteurs de risque, Éd. Modulo-Griffon, Montréal, p 541-551.

Mercer RR, Scabilloni J, Wang L, Kisin E, Murray AR, Schwegler-Berry D, Shvedova AA, Castranova V, 2008. Alteration of depositon pattern and pulmonary response as a result of improved dispersion of aspirated single walled carbon nanotubes in a mouse model. *Am J Physiol Lung Cell Mol Physiol*, 294 : L87–L97.

Michalet X, Pinaud FF, Bentolila LA, Tsay JM, Doose S, Li JJ, Sundaresan G, Wu AM, Gambhir SS, Weiss S, 2005. Quantum Dots for Live Cells, in Vivo Imaging, and Diagnostics. *Science*, 307 (5709) : 538 – 544.

Mills NL, Donaldson K, Hadoke PW, Boon NA, MacNee W, Cassee FR, et al, 2009. Adverse cardiovascular effects of air pollution. *Nat Clin Pract Cardiovasc Med*, 6 : 36-44.

Ministère de la justice, Canada, 2005. Modifications apportées par le projet de loi C-45 aux dispositions du code criminel sur la responsabilité pénale des organisations. Ministère de la justice, Canada, http://canada.justice.gc.ca/fR-Dept/pub/c45/.

Mohlmann C, Welter J, Klenke M, Sander J, 2009. Aerosols at nanomaterial production and handling processes. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Mohlmann C. Occurrence of ultrafine aerosols at the work place, 2005. Vorkommen ultrafeiner Aerosole an Arbeitsplaetzen. *Gefahrstoffe - Reinhaltung der Luft*, 65 : 469-471.

Monteiro-Riviere NA, Inman AO, 2006. Challenges for assessing carbon nanomaterial toxicity to the skin. *Carbon*, 44 : 1070-1078.

Montovani E, Porcari A, Meili C, Widmer M, 2009. Mapping study on regulation and governance of nanotechnologies. FramingNano Report, 138 p.

Muller J, Huaux F, Lison D, 2006. Respiratory toxicity of carbon nanotubes: How worried should we be? *Carbon*, 44 : 1048-1056.

Muller J, Huaux F, Moreau N, Misson P, Heilier JF, Delos M, Arras M, Fonseca A, Nagy JB, Lison D, 2005. Respiratory toxicity of multi-wall carbon nanotubes. *Toxicology and Applied Pharmacology*, 207 : 221-231.

Nasterlack M, Zober A, Oberlinner C, 2008. Considerations on occupational medical surveillance in employees handling nanoparticles *Int Arch Occup Environ Health*, 81 : 721–726.

National Science and Technology Council, 2008. National Nanotechnology Initiative - Strategy for Nanotechnology-Related Environmental, Health and Safety Research, US Government, 102 p.

National Science and Technology Council, Committee on Technology, Subcommittee on Nanoscale Science, Engineering, and Technology, Nanotechnology Environmental and Health Implications Working Group, 2007. Prioritization of Environmental, Health, and Safety Research Needs for Engineered Nanoscale Materials. An interim document for public comment, 8 p. http://www.nano.gov/html/society/ehs_priorities/.

Nemmar A, Hoylaerts MF, Hoet PHM, Vermylen J, Nemery B, 2003. Size effect of intratracheally instilled particles on pulmonary inflammation and vascular thrombosis. *Toxicology and Applied Pharmacology*, 186 : 38-45.

Nemmar A, Hoet PH, Vanquickenborne B, Dinsdale D, Thomeer M, Hoylaerts MF, Vanbilloen H, Mortelmans L, Nemery B, 2002a. Passage of inhaled particles into the blood circulation in humans. *Circulation*, 105 (4) : 411-414.

Nemmar A, Hoylaerts MF, Hoet PHM, Dindsdale D, Smith T, Xu H *et coll.*, 2002b. Ultrafine particles affect experimental thrombosis in an in vivo hamster model. *American Journal of Respiratory and Critical Care Medicine*, 166 : 998-1004.

Nemmar A, Hoet PHM, Thomeer M, Nemery B, Vanquickenborne B, Vanbilloen H, Mortelmans L, Hoylaerts MF, Verbruggen A, Dinsdale D, 2002c. Passage of inhaled particles into blood circulation in humans – Response. *Circulation*, 106 (20) : E141-142.

Nikula KJ, Avila KJ, Griffith WC, Mauderly JL, 1997. Lung tissue responses and sites of particle retention differ between rats and Cynomolgus monkeys exposed chronically to diesel exhaust and coal dust. *Fund Appl Toxicol*, 37 : 37-53.

NIOSH, 2009a. Current Intelligence Bulletin 60. Interim Guidance for Medical Screening and Hazard surveillance for Workers Potentially Exposed to Engineered Nanoparticles. Department of Health and Human Services, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, 40 p.

NIOSH, 2009b. Approaches to Safe Nanotechnology Managing the Health and Safety Concerns Associated with Engineered Nanomaterials. Department of Health and Human Services, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, Publication 2009-125, 104 p.

NIOSH, 2009c. Qualitative risk characterization and management of occupational hazards : control banding (CB). A literature review and critical analysis. Department of Health and Human Services, Center for Disease Control and Prevention, National Institute for Occupational Safety and Health, 118 p.

NIOSH, 2009d. Progress Toward Safe Nanotechnology in the Workplace. A Report from the NIOSH Nanotechnology Research Center, Department of Health and Human Services, Center for Disease Control and Prevention, NIOSH, Nanotechnology Research Program, 166 p.

NIOSH, 2008a. Approaches to Safe Nanotechnology: An Information Exchange with NIOSH. Disponible en ligne à : http://www.cdc.gov/niosh/topics/nanotech/safenano/.

NIOSH, 2008. Strategic plan for NIOSH nanotechnology research and guidance : filling the knowledge gap. Draft, Department of Health and Human Services, Center for Disease Control and Prevention, NIOSH, Nanotechnology Research Program, 103 p.

NIOSH, 2007. Progress Toward Safe Nanotechnology in the Workplace, Rapport du NIOSH Nanotechnology Research Center, Department of Health and Human Services, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, 177 p.

NIOSH, 2006. National Institute for Occupational Safety and Health – Centres for Disease Control and Prevention, Approaches to safe nanotechnology – An information exchange with NIOSH. 37 p.

NIOSH, 2005. NIOSH Current Intelligence Bulletin : Evaluation of Health Hazard and Recommendations for Occupational Exposure to Titanium Dioxide, NIOSH draft, 22 November.

NNI, National Nanotechnology Initiative, 2008. Strategy for nanotechnology-related environmental, health, and safety research, 102 p.

Noël A, Truchon G, 2009. Mécanismes de toxicité pulmonaire *in vitro* des particules ultrafines: synthèse et revue de la littérature, *Travail et santé*, S2-S8.

Obadia A, 2008. Les nanotechnologies: projet d'avis, Conseil économique et social, Paris, 147 p.

Oberdörster G, Stone V, Donaldson K, 2007. Toxicology of nanoparticles: a historical perspective. *Nanotoxicology*, 1 (1) : 2-25.

Oberdörster G, Oberdörster E, Oberdörster J, 2005a. Nanotoxicology: An Emerging Discipline Evolving from Studies of Ultrafine Particles, National Institute of Environmental Health Sciences, EHP, 84 p.

Oberdörster G, 2005b. Inhaled Nano-sized Particles: Potential effects and Mechanisms. Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials, October 12-14 2004, Buxton, UK, p 35-46, Health and Safety Executive and the National Institute for Occupational Safety and Health, United States, July 2005. http://www.hsl.gov.uk/capabilities/nanosymrep_fincollpdf.

Oberdörster G, Maynard A, Donaldson K, Castranova V, Fitzpatrick J, Ausman K, Carter J, Karn B, Kreyling W, Lai D, Olin S, Monteiro-Riviere N, Warheit D, Yang H, 2005c. Principles for characterizing the potential human health effects from exposure to nanomaterials: elements of a screening strategy, *Particle and Fibre Toxicology*, 2 : 8, 35 p.

Oberdörster G, 2004a. Manufactured nanomaterials (Fullerenes, C_{60}) induced oxidative stress in the brain of juvenile largemouth bass. *Environmental health perspectives,* 112 (10) : 1058-1062.

Oberdörster G, 2001. Pulmonary effects of inhaled ultrafine particles, *International Archives of Occupational and Environmental Health*, 74 : 1-8.

Oberdörster G, Finkelstein JN, Johnston C, Gelein R, Cox C, Baggs R *et coll.*, 2000. HEI Research Report : Acute pulmonary effects of ultrafine particles in rats and mice. HEI Research Report No. 96, August, 2000, Health Effects Institute.

Oberdörster G, Gelein R, Ferin J, Weiss B, 1995. Association of particle air pollution and acute mortality: Involvement of ultrafine particles? *Inhalation Toxicology*, 7 : 111-124.

Oberdörster G, Ferin J, Lehnert BE, 1994. Correlation between particle size, in vivo particle persistence, and lung injury. *Environmental Health Perspectives*, 102 (Suppl 5) : 173-9.

Oberdörster G, Ferin J, Gelein R, Soderholm SC, Finkelstein G, 1992. Role of the alveolar macrophage in lung injury: studies with ultrafine particles. *Environmental Health Perspectives*, 97 : 193-199.

Oberdörster G, Ferin J, Finkelstein G, Wade P, Corson N, 1990. Increased pulmonary toxicity of ultrafine particles? II. Lung lavage studies. *Journal of Aerosol Science*, 21 : 384-391.

OCDE, 2009. Emission assessment for the identification of sources and release of airborne manufactured nanomaterials in the workplace : compilation of existing guidance. Environment, Health and Safety Publications, Series on the Safety of Manufactured Nanomaterials, #11. OECD 2009, 25 p.

OCDE, 2008. Environment, Health and Safety Publications, Series on the Safety of Manufactured Nanomaterials Environment Directorate Joint Meeting of the Chemicals Committee and the Working Party on Chemicals, Pesticides and Biotechnology, 2008. Current Developments/ Activities on the Safety of Manufactured Nanomaterials/ Nanotechnologies. Tour de Table at the 2nd Meeting of the Working Party on Manufactured Nanomaterials, Berlin, Germany, 25-27 April 2007 document # ENV/JM/MONO(2007)16, 06-Aug-2007 , 77 p.

Ogilvie Robichaud C, Tanzil D, Weilenmann U, Wiesner WR, 2005. Relative risk analysis of several manufactured nanomaterials: an insurance industry context. *Envir Sci Technol*, 39 : 8985- 8994.

Ono-Ogasawara M, Serita F, Takaya M, 2009. Field survey of workplace handling fullerene. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Ostiguy C, Soucy B, Lapointe G, Woods C, Ménard L, Trottier M, 2008. Health effects of engineered nanoparticles – 2nd Edition. IRSST Report # R-589, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 120 p.

Ostiguy C, Roberge B, Ménard L, Endo CA, 2009. Best Practices Guide to Synthetic Nanoparticle Risk Management. Technical Guide, IRSST Report # R-599, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 73 p.

Ostiguy C, Lapointe G, Ménard L, Cloutier Y, Trottier M, Boutin M, Antoun Monty, Normand C, 2006b. Les nanoparticules : connaissances actuelles sur les risques et les mesures de prévention en santé et en sécurité du travail. IRSST Report # R-455, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 90 p.

Ostiguy C, Lapointe G, Ménard L, Cloutier Y, Trottier M, Boutin M, Antoun M, Normand C, 2006a. Les effets sur la santé reliés aux nanoparticules. IRSST Report # R-451, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 55 p.

Ostiguy C, Asselin P, Malo S, Nadeau D, DeWals P, 2005. Management of Occupational Manganism : Concensus of an Experts' Panel. Série Études et Recherches, IRSST Report # R-417, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 57 p.

Ostiguy C, Malo S, Asselin P, 2003. Synthèse des connaissances scientifiques sur les risques d'atteinte à la santé suite à une exposition professionnelle au manganèse. IRSST Report # R-339, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 57 p.

Ostiguy C, Lesage J, 1998. Les émissions de moteurs diesels : revue sommaire de leur composition et des risques de développement de cancer pulmonaire, IRSST Report # R-194, Institut de recherche Robert-Sauvé en santé et sécurité du travail, Montréal, 32 p.

Paik SY, DM Zalk, P Swuste, 2008. Application of a pilot control banding tool for risk assessment and control of nanoparticle exposures. *Ann Occup Hyg*, 52 (6) : 419-428.

Panessa-Warren BJ, Warren JB, Wong SS, Misewich JA, 2006. Biological cellular response to carbon nanoparticle toxicity. *J Phys : Condens Matter*, 18 : S2185-S2201.

Parlement européen, 2009. Proposition de résolution du parlement européen sur les aspects réglementaires des nanomatériaux, Rapport sur les aspects réglementaires des nanomatériaux, Commission de l'environnement, de la santé publique et de la sécurité alimentaire, 22 p.

Particle measurement Systems, 2009. Particle Measuring Systems introduces the Nano-ID^{TM} – a world-class wide range sampling system for Occupational & Industrial Hygiene air monitoring, http://www.pmeasuring.com/nano.

Pautrat JL, 2003. Demain le nanomonde, Voyage au cœur du minuscule. Paris, Fayard, 250 p.

Pekkanen J, Peters A, Hoek G, Tiittanen P, Brunekreff B, de Hartog J, *et coll.* 2002. Particulate air pollution and risk of ST-segment depression during repeated submaximal exercise tests among subjects with coronary hearth disease. The exposure and risk assessment for fine and ultrafine particles in ambient air (ULTRA) study. *Circulation*, 106 : 933-938.

Pekkanen J, Timonen KL, Ruuskanen J, Reponen A, Mirme A, 1997. Effects of ultrafine and fine particles in urban air on peak expiratory flow among children with asthmatic symptoms. *Environmental Research*, 74 (1) : 24-33.

Penttinen P, Timonen KL, Tiittanen P, Mirme A, Ruuskanen J, Pekkanene J, 2001. Ultrafine particles in urban air and respiratory health among adult asthmatics. *European Respiratory Journal*, 17 (3) : 428-435.

Peters TM, Elzey S, Johnson R, Park H, Grassian VH, Maher T, O'Shaughnessy P, 2009. Airborne monitoring to distinguish engineered nanomaterials from incidental particles for environmental health and safety, *J Occup Env Hyg*, 6 : 73-81.

Peters G, 2005. Risk evaluation & control : Current perspective, Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials : Nanomaterials a risk to health at work ? by David Mark, October 12-14 2004, Buxton, UK, Health and Safety Executive and the National Institute for Occupational Safety and Health, United States, July 2005, p. 84-93.

Peters A, Wichmann HE, Tuch T, Heinrich J, Heyder J, 1997. Respiratory effects are associated with the number of ultrafine particles. *Am Resp Crit Care Med*, 155 : 1376-1383.

Poland CA, Duffin R, Kinloch I, Maynard A, Wallace WAH, Seaton A, Stone V, Brown S, MacNee W et Donaldson K, 2008. Carbon nanotubes introduced into the abdominal cavity of mice show asbestos-like pathogenicity in a pilot study. *Nature Nanotechnology*, 3 : 423-428.

Pope CA, Burnett RT, Thurston GD, Thun MJ, Calle EE, Krewski D, Godleski JJ, 2004. Cardiovascular mortality and long-term exposure to particulate air pollution: epidemiological evidence of general pathophysiological pathways of disease. *Circulation,* 109 (1) : 71–74.

Pope CA, Burnett RT, Thun MJ, Calle EE, Krewski E, Ito K, Thurston GD, 2002. Lung cancer, cardiopulmonary mortality and long term exposure to fine particulate air pollution. *JAMA,* 287 (9) : 1132–1141.

Preining, 1998. The physical nature of very, very small particles and its impact on their behaviour. *Journal of Aerosol Science,* 29 : 481-495.

Pritchard DK, 2004. Literature review – explosion hazards associated with nanopowders, Health and Safety Laboratory, HSL/2004/12, Harpur Hill, Buxton, UK, 22 p.

Proust C, 2005. Le risque d'explosion induit par la manipulation de nanoparticules d'origine organique et minérales dans Agence Nationale de la Recherche, Santé-environnement et santé-travail : nouvelles perspectives de recherches. Séminaire de prospective scientifique et de lancement du programme recherche du Plan national santé environnement et du Plan santé travail, Partie 1.2 Interface milieux/ impacts sur la santé, Ministère délégué à la recherche, République Française, 31 March 1st April 2005, p. 8-9. http://www.recherche.gouv.fr/rapport/santetravail/1.2intermilieu_impactsant%E9.pdf.

Pui DYH, Kim SC, 2006. Penetration of nanoparticles through respirator filter media. Minneapolis, MN: University of Minnesota, Mechanical Engineering Department, Particle Technology Laboratory. NIOSH Contract No. 254–2005–M–11698 for National Personal Protective Technology Division.

Rao CNR, 2004. New developments of nanomaterials. *Journal of Materials Chemistry,* 14 (4) : E4.

Rao CNR, Müller A, Cheetham AK, 2004. *The Chemistry of Nanomaterials. Synthesis, Properties and Applications.* Weinheim, Wiley, 761 p.

Règlement sur la santé et la sécurité du travail [S-2.1, r.19.01], 2007. Québec : Éditeur officiel. http://www.csst.qc.ca/portail/fr/publications/RSST.htm.

Rejeski D, 2008. "National Nanotechnology Initiative: Charting the Course for Reauthorization", présentation au United States Senate Committee on Commerce, Science, and Transportation, Subcommittee on Science, Technology, and Innovation, April 24, 2008.

Rengasamy S, Eimer BC et Shaffer RE, 2009. Comparison of nanoparticles filtration performance of NIOSH-approved and CE-Marked particulate filtering facepiece respirators. *Ann Occup Hyg,* 53 (2) : 117-128.

Rengasamy S, King WP, Eimer BC, Shaffer RE, 2008. Filtration Performance of NIOSH-Approved N95 and P100 Filtering Facepiece Respirators Against 4 to 30 Nanometer-Size Nanoparticles. *J Occ Env Hyg,* 5 : 556–564.

Renwick LC, Brown D, Clouter A, Donaldson K, 2004. Increased inflammation and altered macrophage chemotactic responses caused by two ultrafine particle types. *Occup Environ Med,* 61 : 442-447.

Renwick LC, Donaldson K, Clouter A, 2001. Impairment of Alveolar Macrophage Phagocytosis by Ultrafine Particles. *Toxicology and Applied Pharmacology,* 172 (2) : 119-127.

Richardson AW, Eshbaugh JP, Hofacre KC, Gardner PD, 2006. Respirator filter efficiency testing against particulate and biological aerosols under moderate to high flow rates. ECBC-CR-085. Battelle Memorial Institute, 505 King Avenue, Columbus, OH 43201-2693 for the U.S. Army Edgewood Chemical Biological Center. http://www.cdc.gov/niosh/npptl/researchprojects/pdfs/CR-085Gardner.pdf .

Riediker M, Meyer T, Imhof C, Ferraris G, Schmid K, 2009. Exposure scenarios in research and production involving nanomaterials, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Roberge B, Deadman E, Legris M, Ménard L, Baril M, 2004. Manuel d'hygiène du travail. Du diagnostic à la maîtrise des facteurs de risque, Éd. Modulo-Griffon, 738 p.

Roco MC, 2009. National Nanotechnology Investment in the FY 2009 Budget Request, AAAS Report XXXIII, Research and development FY 2009.

Romieu I, Meneses F, Ruiz S, Sienaaara JJ, Huerta J, White MC, Etzel RA, 1996. Effects of Air pollution on the respiratory health of asthmatic children living in Mexico City. *American Journal of Respiratory and Critical Care Medicine*, 154 : 300-307.

Roszek B, de Jong WH, Geertsma RE, 2005. Nanotechnology in medical applications: state-of-the-art in materials and devices. RIVM report 265001001/2005, 123 p.

Royal Society – Science Council of Japan, 2005. Report of a joint Royal Society – Science Council of Japan workshop on the potential health, environmental and societal impacts of nanotechnologies, 11 and 12 July 2005, 15 p. http://www.royalsoc.ac.uk/displaypagedoc.asp?id=17357.

Royal Society & Royal Academy of Engineering, 2004. Nanoscience & Nanotechnologies – opportunities and uncertainties, Report July 2004. http://www.nanotec.org.uk/finalReport.htm.

Ruckerl R, Ibald-Mulli A, Koenig W, Schneider A, Woelke G, Cyrys J, Heinrich J, Marder V, Frampton M, Wichmann HE, Peters A, 2006. Air pollution and markers of inflammation and coagulation in patients with coronary heart disease. *Am J Respir Crit Care Med*, 173 (4) : 432–441.

Safe Work Australia, 2009a. Engineered Nanomaterials: evidence on the effectiveness of workplace controls to prevent exposure, Safe Work Australia, November 2009, 82 p.

Safe Work Australia, 2009. Engineered Nanomaterials: A review of the toxicology and health hazards, Safe Work Australia, November 2009, 182 p.

Salehi F, 2005. Neurotoxicity and neurobehavioral effects of manganese phosphate/sulfate mixture in male Sprague-Dawley rats following subchronic inhalation exposure. Thèse de doctorat, Université de Montréal, Montréal.

Sandler R, 2009. *Nanotechnology: The Social and Ethical Issues.* Project on Emerging Nanotechnologies, Woodrow Wilson International Center for Scholars or The Pew Charitable Trusts, January 2009, 63 p.

Sano N, Wang H, Alexandrou I, Chhowalla M, Teo KBK, Amaratunga GAJ, Iimura K, 2002. Properties of carbon onions produced by an arc discharge in water. *Journal of Applied Physics*, 92 (5) : 2783-2788.

Santé Canada, Environnement Canada, 2007. Proposition de cadre réglementaire pour les nanomatériaux en vertu de la loi canadienne sur la protection de l'environnement (1999), 10 septembre 2007, http://www.ec.gc.ca/substances/nsb/fra/nanoproposition_f.shtml.

Sayes CM, Liang F, Hudson JL, Mendez J, Guo W, Beach JM, Moore VC, Doyle CD, West JL, Billups WE, Ausman KD, Colvin VL, 2006. Functionalization Density Dependence of Single-Walled Carbon Nanotubes Cytotoxicity in Vitro, *Toxicology Letters*, 161 : 135-142.

Sayes CM, Fortner JD, Guo W, Lyon D, Boyd AM, Ausman KD, Tao YJ, Sitharaman B, Wilson LJ, Hugues JB, West JL, Colvin VL, 2004. The Differential Cytotoxicity of Water-Soluble Fullerenes, *Nano Letters*, 4 (10) : 1881-1887.

SCENIHR, 2009. Risk Assessment of Products of Nanotechnologies, Communauté européenne, 71 p.

SCENIHR, 2007. Opinion on the appropriateness of the risk assessment methodology in accordance with the technical guidance documents for new and existing substances for assessing the risk of nanomaterials. http://ec.europa.eu/health/ph_risk/committees/04_scenihR-Docs/scenihr_o_004c.pdf.

Schiller CF, Gebhart J, Heyder J, Rudolf G, Stahlhofen W, 1988. Deposition of monodisperse insoluble aerosol particles in the 0.0005 to 0.2 µm size range within the human respiratory tract. *Annals of Occupational Hygiene,* 32 (suppl 1) : 41-49.

Schmidt K, Riediker M, 2008. Use of Nanoparticles in Swiss Industry : A Targeted Survey. *Env Sci Technol*, 42 (7) : 2253-2260.

Schnieder T, Jannson A, Jensen KA, Kristjansson V, Luotamo M, Nygren O, Savolainen K, Skaug V, Thomassen Y, Tossavainen A, Tuomi T, Wallin H, 2007. Evaluation and control of occupational health risks from nanoparticles, Nordic Council of Ministers, Copenhagen, TemaNord, 2007 : 581, 96 p.

Schnieder T, Cherrie JW, Vermeulen R, Kromhout H, 2000. Dermal exposure assessment. *Ann Occup Hyg*, 44 (7) : 493-499.

Schneider T, Vermeulen R, Brouwer DH, Cherrie JW, Kromhout H, Fogh CL, 1999. Conceptual model for assessment of dermal exposure. *Occupational and Environmental Medicine*, 56 : 765-773.

Schulte PA, Schubauer-Berigan MK, Mayweather C, Geraci CL, Zumwalde R, McKernan JL, 2009. Issues in the Development of Epidemiologic Studies of Workers Exposed to Engineered Nanoparticles, *JOEM*, 51 (3) : 323-335.

Schulte PA, Salamanca-Buentello F, 2007. Ethical and scientific issues of nanotechnology in the workplace. *Env health Perspec*, 115 (1) : 5-12.

Schuster F, 2007. Trends in European Research on the Risk Assessment of Nanomaterials. Conférence présentée lors du colloque SAPHIR, "A workshop on innovating industrial processes applied to new materials", Sherbrooke, Québec, 1-2 October 2007.

Schwartz J, Dockery DW, Neas LM, 1996. Is daily mortality associated specifically with fine particles? *J Air & Waste Management Association*, 46 : 927-939.

Schwartz J, 1995. Short term fluctuations in air pollution and hospital admissions of the elderly for respiratory disease, *Thorax*, 50 : 531-538.

Schwartz J, Slater D, Larson TV, Pierson WE, Koenig JQ, 1993. Particulate air pollution and hospital emergency room visits for asthma in Seattle. *Am Rev Respir Dis*, 147 : 826-831.

Schwartz J, Spix C, Wichmann HE, Mali E, 1991. Air pollution and acute respiratory illness in five German communities. *Environ Res*, 56 : 1-4.

Shakesheff AJ, 2005. Problems and solutions of current manufacture of nanoparticles, Proceedings of the First International Symposium on Occupational Health Implications of Nanomaterials, October 12-14 2004, Buxton, UK, Health and Safety Executive and the National Institute for Occupational Safety and Health, United States, July 2005, p. 94-102

Shin WG, Pui DYH, Fissan H, Neumann S,. Trampe A, 2007. Calibration and numerical simulation of Nanoparticle Surface Area Monitor, (TSI Model 3550 NSAM) *Journal of Nanoparticle Research*, 9 : 61–69.

Shinohara N, Gamo M, Nakanishi J, 2009a. Risk assessment of manufactured nanomaterials : fullerene (C_{60}), National Institute of Advanced Industrial Science and technology (AIST), 39 p.

Shinohara N, Ogura I, Gamo M, 2009. Exposure to fullerene C_{60} particles during handling in a pilot plant, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Shiohara A, Hoshino A, Hanaki K, Suzuki K, Yamamoto K, 2004. On the cytotoxicity caused by quantum dots. *Microbiol Immunol*, 48 (9) : 669-75.

Shvedova AA, Kisin E, Murray AR, Johnson V, Gorelik O, Arepalli S, Hubbs AF, Mercer RR, Stone S, Frazer D, Chen T, Deye G, Maybnard A, Baron P, Mason R, Kadiiska M, Stadler K, Mouithys-Mickalad A, Castranova V, Kaagan VE, 2008. Inhalation of carbon nanotubes induces oxidative stress and cytokine response causing respiratory impairment and pulmonary fibrosis in mice. *Toxicologist*, 102 : A1497.

Shvedova AA, Kisin ER, Mercer R, Murray AR, Johnson VJ, Potapovich AI, Tyurina YY, Gorelik O, Arepalli S, Schwegler-Berry D, Hubbs AF, Antonini J, Evans DE, Ku BK, Ramsey D, Maynard A, Kagan VE, Castranova V, Baron P, 2005. Unusual inflammatory and fibrogenic pulmonary responses to single-walled carbon nanotubes in mice, *American Journal of Physiology – Lung Cellular and Molecular Physiology*, 289 : 698-708.

Spielvogel , Hagler R, Keck L, Schneider F, Pesch M, Grimm H, 2009. A new portable instrument for real-time monitoring of airborne nanoparticles, *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

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

Stefaniak AB, Hackley VA, Patri A, Postek MT, 2009. Nanoscale reference materials for environmental, health and safety appplications. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

Stone V, Hankin S, Aitken R, Aschberger K et coll., 2009. Engineered nanoparticles : review of health and environmental safety (ENRHES), Edinburg Napier University, 426 p.

Swiss Engineering, 2006. Déceler les risques des nanoparticules. Revue Technique Suisse RTS http://www.swissengineering-stz.ch/pdf/rts001656.pdf.

Tabata Y, Ikada Y, 1988. Effect of the size and surface charge of polymer microspheres on their phagocytosis by macrophages. *Biomaterials,* 9 : 356-362.

Tabet Y, Bussy C, Amara N, Setyan A, Grodet A, Rossi MJ, Pairon J-C, Boczkowski J et Lanone S, 2009. Adverse effects of industrial multiwalled carbon nanotubes on human pulmonary cells, *J Tox Env Health,* 72 : 60-73.

Takagi A, Hirose A, Nishimura T, Fukumori N, Ogata A, Ohashi N, Kitajima S, Kanno J, 2008. Induction of mesothelioma in p53+/- mouse by intraperitoneal application of multi-wall carbon nanotube. *J Toxicol Sci,* 33 (1) : 105–16.

Teague EC, 2004. *Responsible development of nanotechnology National Nanotechnology Initiative Meeting, April 2, 2004.* National Nanotechnology Initiative, 29 p.

Tejral G, Panyala NR, Havel J, 2009. Carbon nanotubes: toxicological impact on human health and environment. *J Applied Biomedecine,* 7 : 1-13.

The Synergist, 2009. EPA issues new use rules (Nanotechnology), 2009. *The Synergist,* August 2009, p. 63.

Thomas D, Mouret G, Calle-Chazelet S, 2008. Filtration des nanoparticules : un problème de taille, *Hygiène et sécurité du travail, Cahier de notes documentaires*, INRS, France, 211 : 8-19.

Timonen KL, G Hoek, J Heinrich, A Bernard, B Brunekreef, J de Hartog, K Hameri, A Ibald-Mulli, A Mirme, A Peters, P Tiittanen, WB Kreyling, J Pekkanen, 2004. Daily variation in fine and ultrafine particulate air pollution and urinary concentrations of lung Clara cell protein CC16. *Occup Environ Med,* 61 (11) : 908–914.

Tjälve H, Henriksson J, 1999. Uptake of metals in the brain via olfactory pathways. *NeuroToxicology,* 20 (2-3) : 181-196.

Tomalia DA, 2004. Birth of a new macromolecular architecture: dendrimers as quantized building blocks for nanoscale synthetic organic chemistry, *Aldrichimica Acta,* 37 (2) : 39-57.

Tran CL, Hankin SM, Ross B, Aitken RJ, Jones AD, Donaldson K, Stone V, Tantra R, 2008. An outline scoping study to determine whether high aspect ratio nanoparticles (HARN) should raise the same concerns as do asbestos fibres. IOM Report on Project CB0406, August 13, 59 p.

Tran CL, Aitken R, Donaldson K, Monteiro-Riviere NA, Oberdörster E, Oberdörster G, Stone V, 2006. Overcoming Obstacles to Effective Research Design in Nanotoxicology, *Proceedings of the Conference* on April 24-25, 2006, Taylor & Francis Group and the Journal of Nanotoxicology, Cambridge/Massachusetts, USA.

Tran CL, Buchanan D, Cullen RT, Searl A, Jones AD, Donaldson K, 2000. Inhalation of poorly soluble particles II. Influence of particle surface area on inflammation and clearance. *Inhalation Toxicology*, 12 : 1113-1126.

Tsai S, Marzik J, Lewis R, Ellenbecker, 2009. An investigation of nanoparticles release at a facility producing boron nanoparticles, 2009. *Proceedings of the 4th International Conference on Nanotechnology Occupational and Environmental Health*, 26-29 August 2009, Helsinki.

TSI, 2009. Scanning Mobility Particle Sizer™ Spectrometers. http://www.tsi.com/en-1033/segments/particle_instruments/2212/scanning_mobility_particle_sizer™_spectrometers.aspx.

Tsuji JS, Maynard AD, Howard PC, James JT, Lam CW, Warheit DB, Santamaria AB, 2006. Research Strategies for Safety Evaluation of Nanomaterials, Part IV: Risk Assessment of Nanoparticles, Forum Series, *Toxicological Sciences*, 89 (1) : 42-50.

US DOE, 2007. Approach to Nanomaterial ES&H; Revision 2-June 2007, U.S. Department of Energy's Nanoscale Science Research Centers. Washington, DC: U.S. Department of Energy.

United States Environmental Protection Agency, 2009. Proposed Significant New Use Rules on Certain Chemical Substances. Federal Register 74 (6 November 2009) : 57430-57436.

United States Environmental Protection Agency, 2008a. Federal Register Environmental Documents/ Vol. 73, No. 212 / Friday, October 31, 2008 / Notices.

United States Environmental Protection Agency, 2008b. Federal Register Environmental Documents / Vol. 73, No. 215 / November 5, 2008 / Notices, p 65743-65766.

United States Environmental Protection Agency, 2007. Nanotechnology white paper. Science Policy Council, U.S. Environmental Protection Agency, Washington, DC 20460, 136 p.

USACHPPM, 2006. OSHA's Final Rule on Assigned Protection Factors for Respirators, Fact Sheet 55-011-1106. Industrial Hygiene Field Services, U.S. Army Center for Health Promotion and Preventive Medicine, 5158 Blackhawk Road, Aberdeen Proving Ground, MD 21010-5403 410-436-3118 or DSN 584-3118, 2006.

VanOsdell DW, Liu BYH, Rubow KL, Pui DYH, 1990. Experimental Study of Submicrometer and Ultrafine Particle Penetration and Pressure Drop for High Efficiency Filters, *Aerosol Science and Technology*, 12 (4) : 911-925.

Viau C, Truchon G, 2004. *Évaluation de l'exposition cutanée, In* Manuel d'hygiène du travail, du diagnostic à la maîtrise des facteurs de risque, Édité par B Roberge, JE Deadman, M Legris, L Ménard et M Baril, Modulo Griffon, Montréal, p 511-524.

Vincent JH, 2005. Health-related aerosol measurement : a review od existing sampling criteria and proposals for new ones. *J Envir Monit,* 7 : 1037-1053.

Vineis P, 2005. Scientific Basis for The Precautionary Principle. *Toxicology and Applied Pharmacology*, 207 (2 Suppl) : S658-S662.

Wake D, 2006. The assessment of different metrics of the concentration of nano (ultrafine) particles in existing and new industries, Health and Safety Laboratory, 80 p.

Wang J, Chen DR, Pui DYH, 2007. Modeling of filtration efficiency of nanoparticles in standard filter media. *Journal of Nanoparticle Research*, 9 : 109-115.

Warheit D, 2009. Assessing the Role of Surface Reactivity Indices in Nanoparticle-related Pulmonary Toxicity, conference présentée au National Research Council Canada, Ottawa, Canada, le 6 février 2009.

Warheit DB, Webb TR, Colvin VL, Reed KL et Sayes CM, 2007. Pulmonary Bioassay Studies with Nanoscale and Fine-Quartz Particles in Rats: Toxicity is Not Dependent upon Particle Size but on Surface Characteristics. *Toxicol Sci,* 95 : 270-280.

Warheit D, Webb T, Sayes C, Colvin V, Reed K, 2006. Pulmonary Instillation Studies with Nanoscale TiO_2 Rods and Dots in Rats: Toxicity is not Dependent Upon Particle Size and Surface Area, *Toxicological Sciences*, 91 : 227-236.

Waters J, 2003. Global Research & Development (R&D) Expenditure on Nanotechnology, Annex to HSC/04/42. [S.l. s.n.], 13 p.

Whitaker J, 2005. Pharmaceutical Dust Extraction and Vacuum Cleaning http://www.ptemag.com/pharmtecheurope/Analytical/Pharmaceutical-Dust-Extraction-and-Vacuum-Cleaning/ArticleStandard/Article/detail/183004?contextCategoryId=39141&ref=25

Wichmann HE, Peters A, 2000. Epidemiological evidence of the effects of ultrafines particles exposure. *Philosophical Trans R Soc*, 358 : no. 1775.

Witschger O, Fabriès JF, 2005. Particules ultrafines et santé au travail: 1- caractéristiques et effets potentiels sur la santé. Hygiène et sécurité du travail. *Cahiers de notes documentaires*, INRS, 199 : 21-35.

Zalk D, Kamerzelli R, Paik S, Kapp J, Harrington D, Swuste P, 2010. Risk Level Based Management System: A Control Banding Model for Occupational Health and Safety Risk Management in a Highly Regulated Environment. *Industrial Health*, 48 (1) : 18-28.

Zhang Z, Kleinstreuer C, Donohue JF, Kim CS, 2005. Comparison of micro- and nano-size particle depositions in a human upper airway model. *Journal of Aerosol Science*, 36 (2) : 211-233.

Zhang Q, Kusaka Y, Donaldson K, 2000. Comparative pulmonary responses caused by exposure to standard cobalt and ultrafine cobalt. *J Occup Health*, 42 : 179-184.

Zhao Y, Meng H, Chen Z, Zhao F, Chai Z, 2007. “Biological Activities of Nanomaterials/ Nanoparticles”, In “Nanotoxicology, Interactions of Nanomaterials with Biological Systems”, Édité par Zhao Y et Nalwa, American Scientific Publishers, p 1-28.

Zhou YM, Zhong CY, Kennedy IM, Leppert VJ, Pinkerton KE, 2003a. Oxidative stress and NFkappaB activation in the lungs of rats: a synergistic interaction between soot and iron particles. *Toxicology and Applied Pharmacology*, 190 : 157-169.

Zhou YM, Zhong CY, Kennedy IM, Pinkerton KE, 2003b. Pulmonary Responses of Acute Exposure to Ultrafine Iron Particles in Healthy Adult Rats. *Environmental Toxicology*, 18 : 227-235.