THE HIND SIGHT OF NANOTECHNOLOGY

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ABSTRACT

Nanotechnology addresses at the atomic, molecular, and macromolecular scale, leading to the controlled manipulation and study of structures and devices with length scales in the 1- to 100-nanometers range, that have unique size, surface tailorability, improved solubility, and multifunctionality.

The "Gold-Rush" indicated by the ever hyping investments in this sector is pressing towards a mammoth market of 1 to 2.95 trillion $ by 2015. Categorization of this market shows that nanomaterials stakes barely 0.5% of the total market size encompassing into nanomaterials, nanointermediates, and nano-enabled products.

But, nanotubes and related market is alone projected to cross US$5 billion by the year 2012. In view of the toxicity that can be posed by it serious problems can threaten to neutralize the gains of nanotechnology.

Research has intimidated experts with several reports on toxicity as nanoparticles can have unrestricted entry into body through inhalation, blood, skin or gut. Oxidative stress is produced as a consequence of the properties of the combustion-derived nano paticles, principally the large surface area, metals, and organic molecules eliciting inflammation. Such stark and aggressive issues are addressed in the paper.

INTRODUCTION

Global Dimming: Is this the first or the last lesson UNLEARNT?

Image above: Sun-blocking aerosols around the world steadily declined since the 1991 eruption of Mount Pinatubo, according to satellite estimates. The decline appears to have brought an end to the "global dimming" earlier in the century (Michael Mishchenko, NASA).
Carbon and sulfate aerosols, or particles, can affect the climate in two ways. The "direct effect" is the scattering and absorption of solar radiation by aerosols. Both sulfate and carbon aerosols scatter light back to space thus acting to reduce the warming caused by "greenhouse" gases. There is also an "indirect effect," in which aerosols affect the reflectivity of clouds, making them"shinier." This also has the opposite effect of reducing global warming by reflecting the sun's heat back into space. Climatologists are now trying to understand the sum of these effects on global climate change.
"Global dimming" phenomenon, is now being admitted to by NASA as a consequence of the haze layers formed by "persistent jet contrails" .

CARBON NANOTUBE - A POSSIBLE PRIMER TO GLOBAL DIMMING !?!

Neolithic Stone Age ice, 10,000 years old showed presence of MWCNTs and fullerene-like nanocrystal forms. (Chiu et al.; 2006) Dr. Smalley predicted that hundreds or thousands of tons of CNTs could be produced in 5 to 10 years (Ball, 2001), and "in time, millions of tonnes of nanotubes will be produced worldwide every year" (Taubes, 2002).

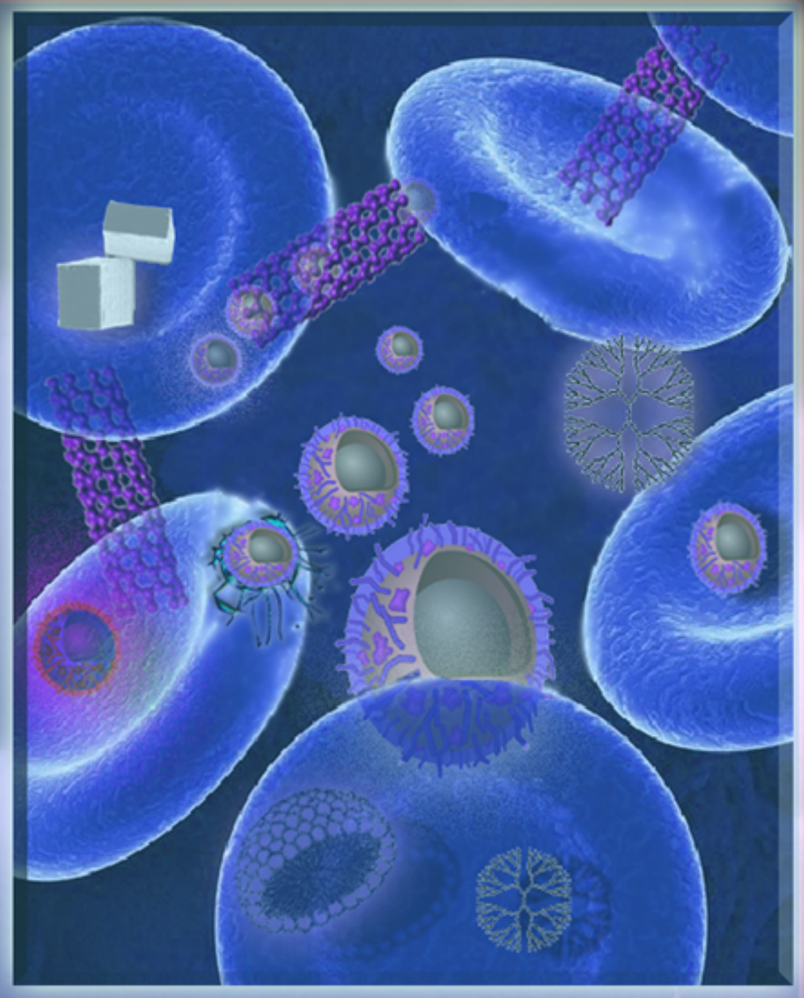
Flames generated by typical kitchen stoves on propane + 96% methane or natural gas gave aggregates of pure carbon or graphene containing several hundred to several thousand MWCNTs (0.4 to 2 µm) or other related nanocrystal forms (20 nm), putting them in the respirable range.
Bang et al. (2004) concluded that MWCNTs and carbonaceous nanoparticles are ubiquitous in the environment; they further speculated that MWCNTs are a major component of indoor and outdoor airborne particulate matter.
Mortality rates in six U.S. cities showed an association between fine particulate air pollution and excess mortality (Dockery et al., 1993) and cardiopulmonary mortality and cardiovascular and pulmonary diseases.
Elemental carbon was shown to have the highest negative correlation ($p = 0.007$) between growth of lung developement of children (10-18). It is noteworthy that the fine (<2.5 µm) samples that contained MWCNTs collected in Houston were from an area in close proximity to a road with heavy traffic.
Upregulation of inflammatory biomarkers was observed only in cells treated with the pollution-derived nanoparticles (Stern & Scot, 2008).

In production ovens, CNTs are made in large amounts, van der Waals forces cause CNTs to have a strong tendency to bundle into microscopic rope like secondary structure in turn, form loose clumps which can become airborne but are mostly larger than respirable size.

Combustion of heterogeneous fuels under varied conditions generate irregular CNTs less orderly, shorter in length, fewer in numbers, and intermingled with other nanoparticles are of respirable size.

The lack of toxicity of carbon black allotrope of CNT highlights the importance of particle characteristics, primarily due to the surface properties and aspect ratio of CNTs, rather than their nanosize.

Experimental studies in rodents have demonstrated that instillation of multiwalled and single-walled CNTs can cause pulmonary inflammation, granulomas, fibrosis, and even death. The fibrosis distant to the granulomatous lesions was presumed to be in response to either single or less-aggregated particles (Shvedova et al., 2005).



CNTs have been shown to induce **platelet aggregation** in vitro and enhance thrombosis in vivo. Inflammatory potency was best correlated not with particle size or surface area, but with surface reactivity as measured by hemolytic potential. In this study, the smallest, 12 nm, quartz particle was less inflammogenic on a mass basis than the largest, 500-nm particle. A previous study examining the inflammatory potency of micronsized quartz particles in rats also found **hemolytic potential** to correlate well with in vivo inflammation.

The discrepancy between the in vitro and in vivo findings may be due to limited dermal penetration in vivo. Subcutaneous implantation of carbon nanotubes or "hat-stacked" carbon nanofibers in rats resulted in a more severe foreign body granuloma-like response.

All CNT samples tested, regardless of the type and amount of metal impurities they contained, induced dose-dependent lesions characterized chiefly by interstitial granulomas in the lungs of mice (0, 3.3, or 17 mg/kg; 7 and 90 days) with increase in lymphocytes and fibrosis. CNT sample contain nickel, which is highly toxic.1mg of a CNT sample showed focally severe foreign-body reactions characterized by widespread prominent foci of particle-laden macrophages and giant cells, and prominent peribronchial and perivascular lymphocytic infiltrates in some animals.

Bronchoalveolar lavage fluid (BALF) showed dose-dependent increases of inflammation, oxidative stress, or cytotoxicity biomarkers in the lungs like total protein concentration, cell counts, concentration of TGF-β, LDH, and γ -glutamyltranspeptidase activities, and glutathione depletion.at (0.5, 1, or 2 mg/kg).

Dose-dependent damage to the mitochondrial DNA in the aortas at 7, 28, and 60 days after CNT was seen (Li et al., 2005) due to altered expression of inflammatory genes (MCP-1 and VCAM-1, in the heart). Hypercholesterolemic (ApoE-/-) mice witnessed increase in atherosclerotic lesions in the brachiocephalic arteries (20 µg SWCNTs; once every 8 weeks).

Raw HiPco CNTs containing 30% iron attribute to oxidation or peroxidation in cells, the authors attributed these oxidative effects to the iron in the carbon nanotubes (Shvedova et al., 2003). Shvedova et al. (2005) found iron-free
CNTs were capable of producing these cytotoxic effects as well in human epidermal keratinocytes. MWCNT 0.4-mg/ml culture were found to release the proinflammatory cytokine IL-8 in a time-dependent fashion.

MWCNT 1 mg/m3 had decreased NK cell function and exposures to 0.3 mg/m3 and higher particle concentrations caused systemic immunosuppression only after 14 days, characterized by reduced T-cell–dependent antibody response to sheep erythrocytes as well as T-cell proliferative ability in presence of mitogen, Concanavalin A. No changes in gene expression were observed in lung; however, interleukin-10 (IL-10) and NAD(P)H oxidoreductase 1 mRNA levels were increased in spleen (Leah et al.; 2007).

In subcutaneous injection model and thereafter in an intranasal model of sensitization with allergen ovalbumin, SWCNT and MWCNT increased serum levels of OVA-specific IgE, the number of eosinophils in BALF and the secretion of Th2-associated cytokines in the mediastinal lymph node.
Only mwCNT and ufCBP with OVA increased IgG2a levels, neutrophil cell numbers and TNF- and MCP-1 levels in BALF, as well as the acute influx of neutrophils after exposure to the particles alone (Nygaard et al; 2007).

NANO-PARTICLES

The spray planes are filling our atmosphere with nano particles of barium on an almost daily basis. Carnicom estimates that barium levels worldwide are eight times higher than acceptable safety limits. The inhaled nano-particles, 80,000 times smaller than the width of a human hair, stick to the moist lung walls and are small enough to easily be absorbed directly into the blood stream where they will gain access to internal organs such as the liver and brain. Since the aerosol operations began in earnest in about 1998 our bodies have been loading with these particulates and we are just now beginning to feel the ill effects (Carnicom.com).

Silver nanoparticles

Products Like:	Kitchen and Bath Paint	Baby Sunscreen	Laptops
	Clothes	Food storage systems	Wet wipes
	Waxes	Cosmetics	

It is claimed to kill pathogens, but is non-selective and kills several metabolically, ecologically and environmentally important microorganisms. The zebrafish embryos exposed to silver nanoparticles showed deposition of particles in vital organs such as the brain and exhibited severe developmental defects

ZnO

Research showed that zinc oxide nanoparticles at certain concentrations got through ryegrass root surfaces, damaged root tissues, entered root cells, and inhibited seedling growth. You can see it in the image here. "A" represents a normal, healthy root tip. But "B" vvshow the root tips in the presences of zinc oxide. The rootcaps are broken, cortical cells are collapsed and the vascular structures have shrunk (umass.edu/psis/personnel/xing.html). ZnO nm (10 nm) at 12 ppm causes apoptosis of neural stem cell within 24 h. (X iaoyong D, Qixia L, Wenting C, Yanli W, Minghong W, Haijiao Z and Zheng J. Nanosized zinc oxide particles induce neural stem cell apoptosis. Nanotechnology 20 (2009) 115101 (7pp).)

It is estimated that the average person consumes 1012 submicron-sized particles per day in a normal diet as a result of food additives consisting primarily of titanium dioxide (TiO_2) and aluminosilicates (Lomer et al., 2002). Incidental nanoparticles are also found in such common sources as wood smoke, and automobile and furnace exhaust (Barregard et al., 2006; Chang et al., 2004; Fang et al., 2005). Levels of incidental nanoparticles in the outdoor environment near heavy traffic areas can range from 5000 to 3,000,000 particles/cm3! (Utell and Frampton, 2000)..

Manganese nanoparticles to induce adverse effects in neuronal cells, including loss of cell viability, induction of oxidative stress, and dopamine depletion

Warheit et al. demonstrated that intratracheal instillation of rats with 1 or 5 mg/kg of 300-nm pigment-grade TiO_2 particles, 200 nm 3 35 nm TiO_2 rods, or 10-nm TiO_2 dots all resulted in equivalent pulmonary inflammation and tissue injury at 24h postexposure.

In any event, since coating of nanoscale TiO_2 particles has been shown to stabilize the particles and prevent photocatalysis, the proper coating of commercial TiO_2 may eliminate any concerns over dermal carcinogenesis from topical application (Stern & Scott; 2007).

Quartz particles (5 mg/kg) produced dose-dependent inflammatory responses, concomitant with foamy alveolar macrophage accumulation and lung tissue thickening but carbonyl iron or graphite particles produced no significant adverse effects (Chiu et al.; 2006).

Nanoparticles from the GI tract is absorbed as per their size (Hillyer and Albrecht, 2001) and surface characteristics with the smaller, hydrophobic, neutral particles having increased absorption. Polystyrene nanoparticles in rats, showed size dependent absorption (50 nm > 100 nm > 300–3000 nm) and confined to the Peyer's patches of the gut associated lymphoid tissue.

The toxicity of cationic dendrimers, for example, appears to be related not to oxidative stress generation, but to disruption of cell membrane integrity through interaction of the positively charged dendrimer terminal groups and the anionic lipids composing the cell membrane (Stern & Scott; 2007).

BIOCOMPATIBILITY IS TO BE ADDRESSED

Silica particle are known to rage inflammatory response in macrophages. LC50 ranged from 20 µg/ml for 7 nm to 592 µg/ml for 300-nm amorphous silica particles against macrophages, but crystalline silica, mean diameter 1.7µM was 253 µg/ml. LC50 for cytotoxicity for all particle sizes based on total administered particle surface area was 85 cm2/ml. they also increased protein secretion of G-CSF(compared to control), TNF-?, RANTES, TNF-, G-CSF, and VEGF. Transcriptome of Cxcl2 (also known as MIP-2), a C-X-C chemokine that exhibits potent neutrophil chemotactic activity in rat lung was induced by over 30-fold also seen in quartz exposure. Others induced were Ccl4 (MIP-1 β) and Ccl3 (MIP-1?), Cxcr4 (4x) and TNF. Except DNA dependent transcription, cell cycle, inflammatory response, apoptosis, morphogenesis, differentiation and signal transduction 500 nm was more responsive than 10 nm.

Whole genome microarray analysis of the early gene expression changes induced by 10- and 500-nm particles showed that the magnitude of change for the majority of genes affected correlated more tightly with particle surface area than either particle mass or number (Waters et al.; 2009).
But Magnetic nanoparticles that enable drug development, protein detection, and gene delivery were found to be biocompatible when coated with silica. Silica-overcoated magnetic nanoparticles containing rhodamine B isothiocyanate (RITC) within silica shell of controllable thickness [MNPs@SiO2(RITC); 50-nm] given ip were found in brain, lung, heart, liver, spleen, kidney, uterus and testis without any apparent toxicity (Kim et al.; 2006).

STEPS TO CURB THE GROWING DANGER

The House of Representatives passed legislation that makes the importance of understanding the possible risks posed by engineered nanomaterials and growing need to learn more about the possible environmental, health and safety dangers posed by some nanoscale materials (National Nanotechnology Initiative Amendments Act of 2009 (H.R. 554)) sponsored by $800 billion stimulus package .
1. Exposure assessment of manufactured nanoparticles.
2. Toxicology of manufactured nanoparticles.
3. Ability to extrapolate manufactured nanoparticle toxicity using existing particle and fiber toxicological databases.
4. Environmental and biological fate, transport, persistence, and transformation of manufactured nanoparticles; and
5. Recyclability and overall sustainability of manufactured nanomaterials. (Dreher, 2004)

Cosmetic products are not subject to premarket approval by the FDA. Thus cosmetic ingredient, including nanoparticles, should be restricted legally from selling and marking those drugs.
Pan-nano-manufacturers should implicate outcomes of EU Nanomaterial projects (10.6 million).
Nanotox / Impart / Nanosafe2 / Particle-Risk / Antistorm.

CONCLUSION

The surge in quantum of processes and products engaging carbon nanotubes and their hybrids with other nanoparticles need more grave and transparent approach in order to ensure the safety of those who are working with them. The end of all disposed products will contaminate the environment, and pose threats. Greater in depth research is needed to unravel the mechanism of these adverse effects and optimize the measures required to abte and control their prevelence in the air in order to protect those that are exposed to this new material.

REFERENCES

Agins, T. Trends (A special report). The Wall Street Journal. 22November.Anvalay, J. Get smart. AATCC Rev. 5, 34–36. (2005).
Ball, P. Roll up for the revolution. Nature, 414, 142. http://www.pbys.msubington.edu/%nbden/P600/Philip Ball tube news.pdf, accessed May 25, 2005.
Bang, J.J., Guerrero, P.A., Lopez, D.A., Murr, L.E., and Esquivel, E.V. (2004).
Carbon nanotubes and other fullerene nanocrystals in domestic propane and natural gas combustion streams. J. Nanosci. Nanotechnol.,4(7):716.
Cecilie U., Stiland H., Samuelsen M., Torunn A., and Lavik M.,Single-walled and Multi-walled Carbon Nanotubes Promote Allergic Immune Responses in Mice. Toxicological Sciences, 10.1093/toxsci/kfp057.
Chiu-wing L., John T. J., Richard M., Stvarsen A., Robert L. H.,A Review of Carbon Nanotube Toxicity and Assessment of Potential Occupational and Environmental Health Risks. Critical Reviews in Toxicology, 36:189–217, 2006
Jun S.K., Tae-Jong Y., Kyeong N.Y., Byung G. K., Sung J. P., Hyun W.K., Kee H. L., Seung B. P., Jin-Kyu L., and Myung H.C., Toxicity and Tissue Distribution of Magnetic Nanoparticles in Mice. TOXICOLOGICAL SCIENCES 89(1), 338–347 (2006)
Katrina M. W., Masiello M.L., Zanger R.C., Barbara J. T, Norman J. K, Ryan D. Q., Bandyopadhyay S., Teeguarden J.G., Pounds J.G., and Brian D. T., Macrophage Responses to Silica Nanoparticles are Highly Conserved Across Particle Sizes. Toxicological Sciences 2009 107(1):553-569
Ken D., Robert A., Lang T., Vicki S., Rodger D., Gavin F., and Alexander A.,Carbon Nanotubes: A Review of Their Properties in Relation to Pulmonary Toxicology and Workplace Safety. TOXICOLOGICAL SCIENCES 92(1), 5–22 (2006).
Kevin L. D., Health and Environmental Impact of Nanotechnology:
Toxicological Assessment of Manufactured Nanoparticles. TOXICOLOGICAL SCIENCES 77, 3–5 (2004).
Leah A. M., Jun G., Randy V., Gigliotti A., Burchiel S. W., McDonald J., Pulmonary and Systemic Immune Response to Inhaled Multiwalled Carbon Nanotubes. TOXICOLOGICAL SCIENCES 100(1), 203–214 (2007).
Li, Z., Salmen, R., Huldermen, T., Klein, R., Shvedova, A., Lorter, M.I., and Simeonova, P.P. Pulmonary exposure to carbon nanotubes induces vascular toxicity. The Toxicologist CD—An Official Journal of the Society of Toxicology, 84 (S-1), 213 (abstr. 1045). (2005)
Lomer, M. C., Thompson, R. P., and Powell, J. J. Fine and ultrafine particles of the diet: Influence on the mucosal immune response and association with Crohn's disease. Proc. Nutr. Soc. 61, 123–130. (2002)
McDonald J., Mitchell L., TOXICOLOGICAL SCIENCES 100(1), (2008) 181–182
Shvedova, A.A., Castranova, V., Kisin, E.R., Schwegler-Berry, D., Murray, A.R., Gandelsman,V.Z., Maynard, A., and Baron, P., Exposure to carbon nanotube material: Assessment of nanotube cytotoxicity using human keratinocyte cells. J. Toxicol. Environ. Health A 66:1909.
Shvedova, A.A., Kisin, E.R., Mercer, R., Murray, A.R., Johnson, V.J., Potapovich, A.I., Tyurina, Y.Y., Gorelik, O., Arepalli, S.,
Schwegler-Berry, D., Hubbs, A.F., Antonini, J., Evans, D.E., Ku, B.K., Ramsey, D., Maynard, A., Kagan, V.E., Castranova, V., and Baron, P. (2005). Unusual inflammatory and fibrogenic pulmonary responses to single-walled carbon nanotubes in mice. Am. J. Physiol.Lung Cell. Mol. Physiol.
Stephan T. S., and Scott E. M.,Nanotechnology Safety Concerns Revisited. TOXICOLOGICAL SCIENCES 101(1), 1–21 (2008)
Taubes, G. Nanotechnology. An interview with Dr. Richard Smalley. ISI Essential Science Indicators Special Topics. http://www.esi-topics.com/nano/interviews/Richard-Smalley.html, accessed May 25, 2005.
Utell, M. J., and Frampton, M. W., Acute health effects of ambient air pollution: The ultrafine particle hypothesis. J. Aerosol Med. 13, 355–359. (2000)
Warheit,D. B. , Laurence B. R., Reed K. L. Reed, Roach D. H. , Reynolds G. A. and Webb. T. R. , Comparative Pulmonary Toxicity Assessment of Single-wall Carbon Nanotubes in Rats. TOXICOLOGICAL SCIENCES 77, 117–125 (2004).
X iaoyong D, Qixia L, Wenting C, Yanli W, Minghong W, Haijiao Z and Zheng J. Nanosized zinc oxide particles induce neural stem cell apoptosis. Nanotechnology 20 (2009) 115101 (7pp).