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Review Article

Mechanisms of nanotoxicity: Generation of reactive oxygen species☆

Peter P. Fu [a,*], *Qingsu Xia* [a], *Huey-Min Hwang* [b], *Paresh C. Ray* [c], *Hongtao Yu* [c]

[a] *National Center for Toxicological Research, US Food and Drug Administration, Jefferson, AR 72079, USA*
[b] *Department of Biology, Jackson State University, Jackson, MS 39217, USA*
[c] *Department of Chemistry and Biochemistry, Jackson State University, Jackson, MS 39217, USA*

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ABSTRACT

Nanotechnology is a rapidly developing field in the 21st century, and the commercial use of nanomaterials for novel applications is increasing exponentially. To date, the scientific basis for the cytotoxicity and genotoxicity of most manufactured nanomaterials are not understood. The mechanisms underlying the toxicity of nanomaterials have recently been studied intensively. An important mechanism of nanotoxicity is the generation of reactive oxygen species (ROS). Overproduction of ROS can induce oxidative stress, resulting in cells failing to maintain normal physiological redox-regulated functions. This in turn leads to DNA damage, unregulated cell signaling, change in cell motility, cytotoxicity, apoptosis, and cancer initiation. There are critical determinants that can affect the generation of ROS. These critical determinants, discussed briefly here, include: size, shape, particle surface, surface positive charges, surface-containing groups, particle dissolution, metal ion release from nanometals and nanometal oxides, UV light activation, aggregation, mode of interaction with cells, inflammation, and pH of the medium.

1. Introduction

Nanomaterials are chemical molecules that have surfaces with at least one dimension smaller than 100 nm; engineered nanomaterials have the same specific physicochemical characteristics and are manufactured intentionally [1]. Nanotechnology is a rapidly developing field in the 21st century. The various commercial uses of nanomaterials for novel applications are increasing exponentially. Nanomaterials are extremely small in size and possess a large surface area per unit of volume. These novel physical characteristics of nanomaterials can result in their having drastically different chemical and biological properties compared to the same material in bulk form. The unique chemical and biological properties of nanomaterials make them useful in many products for humans, including some in industry, agriculture, business, medicine, clothing, cosmetics, and food [1–9].

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* *Corresponding author*. National Center for Toxicological Research, US Food and Drug Administration, 3900 NCTR Road, Jefferson, AR 72079, USA.
E-mail address: peter.fu@fda.hhs.gov (P.P. Fu).

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Nanotechnology research and development by industry and governments worldwide have been increasing dramatically. It has been estimated that by 2015, nanoproducts will contribute approximately $1 trillion to the global economy [3,10,11].

Humans may be exposed to nanomaterials through inhalation (respiratory tract), skin contact, ingestion, and injection (blood circulation) [4]. The tiny size of nanomaterials allows them to pass more easily through cell membranes and other biological barriers, therefore, nanomaterials can be easily taken up into living organisms and cause cellular dysfunction [10,11]. In addition, because of their unique properties, including high surface-to-volume ratios, nanomaterials are reactive or catalytic, and thus can be potentially toxic. For the safe development of nanotechnology and the safe use of commercial nanomaterials, investigations regarding the cellular toxicity and phototoxicity of nanomaterials are needed.

Although the unique properties of nanomaterials have resulted in an exponential increase in their use, cytotoxic and genotoxic data for most manufactured nanomaterials have not been published at a correspondingly high rate [1,4–6,12,13]. Many nanomaterials synthesized as commercial products are introduced daily into our lives. For example, zinc oxide nanoparticles (nano-ZnO) are one of the most commonly used nanomaterials, with industrial and commercial applications, including personal skin and hair care products, sunscreens, pigments, coatings, ceramic products, and paints [2,14–16]. Another example is titanium dioxide nanoparticles (nano-TiO_2), which are among the top nanomaterials, and are widely used as food additives and drug delivery agents in personal care products, paints, plastics, and cosmetics [6,17,18]. The potential harm of nano-ZnO and nano-TiO_2 to human health has attracted public attention. Understandably, the matter of safety and toxicity of nanomaterials has become an issue of interest to the public. Therefore, understanding the interactions of nanomaterials with biological systems is a particularly important scientific issue.

2. Toxicity of nanomaterials

The range of nanotechnology products is wide and they can be classified into several different compound categories, including metals, metal oxides, carbon, silica, and semiconductor nanomaterials [6]. The toxicity of nanomaterials has been studied in different biological systems, both in cell line systems and different organisms, which include rodents, humans, and aquatic species, such as zebrafish [1,19–25], catfish [26], algae [27], and macrophages [28]. Carbon and metallic nanomaterials are among the most widely used types of engineered nanomaterials. Nano-metals, such as nano-gold (nano-Au), nano-silver (nano-Ag), nano-copper, nano-aluminum, nano-nickel, nano-cobalt, and other nanoparticles, have also been extensively studied. Metal nanoparticles are important industrial materials that are widely used as additives in cosmetics, pharmaceuticals, and food colorants [6]. The skin can be exposed to solid nanoparticles through the application of lotions or creams that contain nano-TiO_2 or nano-ZnO as a sunscreen component or fibrous materials coated with nanoscale substances for water or stain-repellent properties. In addition to exposure due to use of consumer products, the manufacture and use of nanoparticles inevitably leads to increased occupational and environmental exposure. The toxicity of metal oxide nanoparticles, such as nano-TiO_2, nano-ZnO, nano-CuO, nano-CuZn, nano-Fe_3O_4, and nano-Fe_2O_3, with nano-TiO_2, nano-ZnO in particular, has been reported [6,12,29–31]. As expected, different nanomaterials exhibit different toxic potency. For example, Zhu et al [32] compared the toxicity of three nano-metal oxides, nano-CuO, nano-CdO, and nano-TiO_2. Nano-CuO was determined to be the most potent in cytotoxicity and DNA damage, leading to 8-hydroxy-2′-deoxysuanosine (8-OHdG) formation, while nano-$TiO_2$was the least, without inducing a significant level of 8-OHdG [32].

The production of carbon nanotubes (CNTs) and graphene oxide is becoming commercially important. Under some experimental conditions, investigators have found that CNTs and graphene oxide are toxic [33–38].

The mechanisms underlying the toxicity of nanomaterials have recently been studied intensively. An important mechanism of nanotoxicity is the generation of reactive oxygen species (ROS), resulting in the subsequent formation of oxidative stress in tissues [1].

3. Overproduction of ROS and cell damage

In the mitochondria of cells, ATP is synthesized by reduction of molecular oxygen to water through a sequence of coupled proton and electron transfer reactions. During this process, a small percentage of the oxygen is not reduced completely, resulting in the formation of superoxide anion radicals, and subsequently other oxygen-containing radicals. Thus, ROS are byproducts of cellular oxidative metabolism, much of which occurs in the mitochondria. Biologically relevant ROS include superoxide anion radicals, hydroxyl radicals, singlet oxygen, and hydrogen peroxide (H_2O_2) [39]. ROS play beneficial physiological roles in cellular signaling systems and induction of mitogenic responses [40,41]. Besides cellular oxidative stress, there are several other biological reactions that can generate ROS *in vivo*. Transition metals such as copper and iron can also participate in one-electron oxidation–reduction reactions, leading to the formation of ROS [42].

Overproduction of ROS can induce oxidative stress, resulting in cells failing to maintain normal physiological redox-regulated functions [43,44]. The damage in cell function and development includes oxidative modification of proteins to generate protein radicals [45], initiation of lipid peroxidation [46–48], DNA-strand breaks, modification to nucleic acids [49], modulation of gene expression through activation of redox-sensitive transcription factors [50,51], and modulation of inflammatory responses through signal transduction [52], leading to cell death and genotoxic effects [3,53–57].

It has been demonstrated that ROS and oxidative stress are associated with many age-related degenerative diseases [41,45,58,59], including amyotrophic lateral sclerosis, arthritis, cardiovascular disease, inflammation, Alzheimer's disease, Parkinson's disease, diabetes, and cancer [8,40,43,52,60–64].

In nuclear and mitochondrial DNA, 8-OHdG is a predominant form of free-radical-induced oxidative lesion. 8-OHdG has been