Applications and Safety of Nanomaterials Used in the Food Industry

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A survey by the United States Food and Drug Administration (USFDA), in 2008, has revealed that more than 80 nanomaterial-containing food and drink items were available for consumption around the world at that time, and estimated that the Japanese market for nanomaterial-containing food could reach 250 billion yen in 2030. Potential uses of nanotechnology have been identified in almost all segments of the industry, including the following four key areas: agriculture, nutritional supplements and nutraceuticals, food processing, and food packaging. Despite a rapid development of its use in the food sector, little is known about the *in vivo* and *in vitro* kinetics of nanomaterials. Consequently, the risks of nanomaterials have not been analyzed. Nanoparticles are reported to be absorbed across the intestinal barrier via transcellular, paracellular, and junctional pathways, but the bioavailability of each material may be different due to different effects of various factors. Questions about these compounds already raised safety concerns, although the history of their use in the food sector is yet short. In this review, we overview the currently available information regarding the safety of the three main nanomaterials used in the food industry to provide the scientific basis for the risk assessments that are necessary for the development and safe use of nanomaterials in the food sector.

Key words: food sector, nanomaterial-containing foods, nanotechnology, safety concern

1. Introduction

Recent innovations in science and technology, as well as their globalization, have brought sweeping changes to people's eating habits. The number and diversity of chemicals in our food, such as additives, agricultural chemical residues, contaminants, as well as new materials and ingredients to maintain or improve health or prevent lifestyle-related diseases, have increased rapidly. We ingest such chemicals throughout our lifetime often without being aware of them.

Because consumer's interests in food safety concerns are increasing, the safety assessment of these chemicals in our food is essential.

Nanomaterials may offer particular cause for concern due to their small size imparting their unique properties. Nanomaterials with diameters of ≤100 nm are currently used in various applications, including food production (for example, to improve food texture). As the size of particles decreases, their specific surface area increases, and the high specific surface area of nanomaterials results in a dramatic increase in surface coverage and water absorbability compared with those of materials with diameters of >100 nm.

In 2015, a consumer products inventory compiled by the Woodrow Wilson International Center for Scholars listed 117 nanomaterial-containing products in the food and beverage category, which includes foods, beverages, and cooking items[1]. It was estimated that the Japanese market for nanomaterial-containing food could reach 250 billion yen in 2030[2]. With the expansion of this market, the likelihood of human exposure to nanoparticles via the intestinal tract will increase. Even though the amounts of nanomaterials present in food are small, they will tend to be consumed over a long period of time, and therefore it is necessary that biological responses to oral ingestion of nanomaterials be assessed and that information about exposure levels be provided. Although there have been a few studies on the biological effects of nanomaterials on humans, and unexpected biological responses to nanomaterials have recently been reported[3,4], the safety of nanomaterials has not been thoroughly assessed. Little is known about their *in vivo* and *in vitro* kinetics at biologically relevant concentrations via different administration routes. In general, chemical risk analysis involves the integration of hazard information and information about absorption, distribution, metabolism, and excretion for the actual exposure route. Nanomaterials, including those contained in foods, are no exception. Owing to the size of nanomaterials that is the same order of magnitude as that of viruses, their toxicokinetics *in vivo* is unpredictable. In addition, the fact that they can penetrate the intestinal barrier and be absorbed into cells must be considered in evaluations of their *in vivo* kinetics.

The aim of this review is to describe the current uses of nanomaterials in the food sector and summarize an overview of the available data on the safety of nanomaterials.

2. Nanotechnologies in the Food Sector

From many reports and reviews on the current and short-term projected applications of nanotechnologies in the food industry, their potential uses are found for almost all segments of the industry[4,5].

Nanotechnologies have been successfully applied to agri-food production, as has been focused by several key reports[6,7]. Application of nanotechnologies in this food sector has begun to grow now since it has been recognized that conventional agricultural techniques cannot further increase productivity or restore damaged ecosystems[6]. New tools were developed also for nanobiosensing applications[7] which can be used for sensing a wide variety of fertilizers and pesticides, as well as moisture. These are expected to be useful for the identification of crop diseases and agrochemical residues[7], the augmentation of agricultural productivity through genetic improvements of plants[8], and the improvement of the ability of plants to absorb nutrients[9]. Consumer's demand for healthy foods has encouraged researchers to apply nanotechnology to food and nutrition. For instance, nanomaterials have been used to coat food packaging, and nanosieves are used to filter out microorganism and even viruses. In addition, the development of analytical methods for assessment of nanotechnology has been remarkable[10,11]. Moreover, application of nanotechnology in the food and nutrition sector has resulted in the development of food ingredients with increased functionality such as improved physical or chemical properties and physiological performance[12].

Such ingredients are widely used to improve the taste and nutritional value of food products and to prolong shelf-life. Many food products containing engineered nanomaterials are now widely available; for example, titanium dioxide and silica nanoparticles are used as food additives[5]. The strong antimicrobial properties of chitosan[13], silver nanoparticles[14], and photocatalytic titanium dioxide[15] have also been exploited in the food sector. In addition, nano-encapsulation technologies are being used for the development of artificial colorants, preservatives, and aroma compounds. These technologies hopefully will make it possible to disperse or solubilize materials that are otherwise poorly soluble in water, to improve the efficacy of additives, and to improve the absorption rate of encapsulated supplements and nutrients.

Food-packaging materials represent the largest category of applications of nanotechnology in the food sector[16]. Containers and wrapping materials that come into direct contact with food contain nanomaterials to improve packaging properties, such as gas impermeability[17] and against temperature and moisture[18]. Nanomaterials can also impart antibacterial activity or carry out deoxygenation reactions. Containers and wrapping materials containing nanosensors,

referred to as intelligent food packaging, have been used to detect the condition of food inside packaging[19]. In Europe, carbon black and silica dioxide nanomaterials have been already used as food contact materials.

3. Behavior of Ingested Nanomaterials

We are intentionally and unintentionally exposed to various substances through dermal, intraoral, and pulmonary routes; and oral absorption is the major route by which we take in chemicals, as well as nutrients and water. Given that nanoparticles are now included in various food products, it is assumed that the digestive organs, including the intestinal tract, are exposed to nanoparticles daily. Nanoparticles ingested orally travel from the mouth to the stomach and intestines. The epithelial cells lining the small intestine absorb most nutrients, as well as nanomaterials, by endocytosis or by diffusion and transport them across the epithelium. As nanoparticles move through the small intestine, they come into contact with intestinal epithelial cells.

Absorption of nanoparticles has been studied using Caco-2 cell monolayers as gut models. These studies have shown that nanoparticles are transported from the luminal side to the basal luminal side of intestinal epithelial cells, probably by endocytosis or transcytosis[20–22]. Several pathways are available for the absorption of particles across intestinal barriers (**Figure 1**). The transcellular route is the most relevant to the uptake of nanomaterials[23] because the tight junctions of epithelial cells eliminate macromolecules[24]. The size limit of the paracellular route, which depends on the animal species and the tissue examined, has been variously reported as being from 0.6 to 5 nm[25]. Some nanomaterials, such as chitosan nanoparticles, were reported to be able to loosen epithelial tight junctions, suggesting that these nanoparticles may promote the paracellular transport of macromolecules[26,27].

Microparticles (>100 nm) in the gut are absorbed mainly through M-cells[28]. M-cells are present in the epithelium of mucosa-associated lymphoid tissues and can transport macromolecules and particles in the gut lumen into cells of
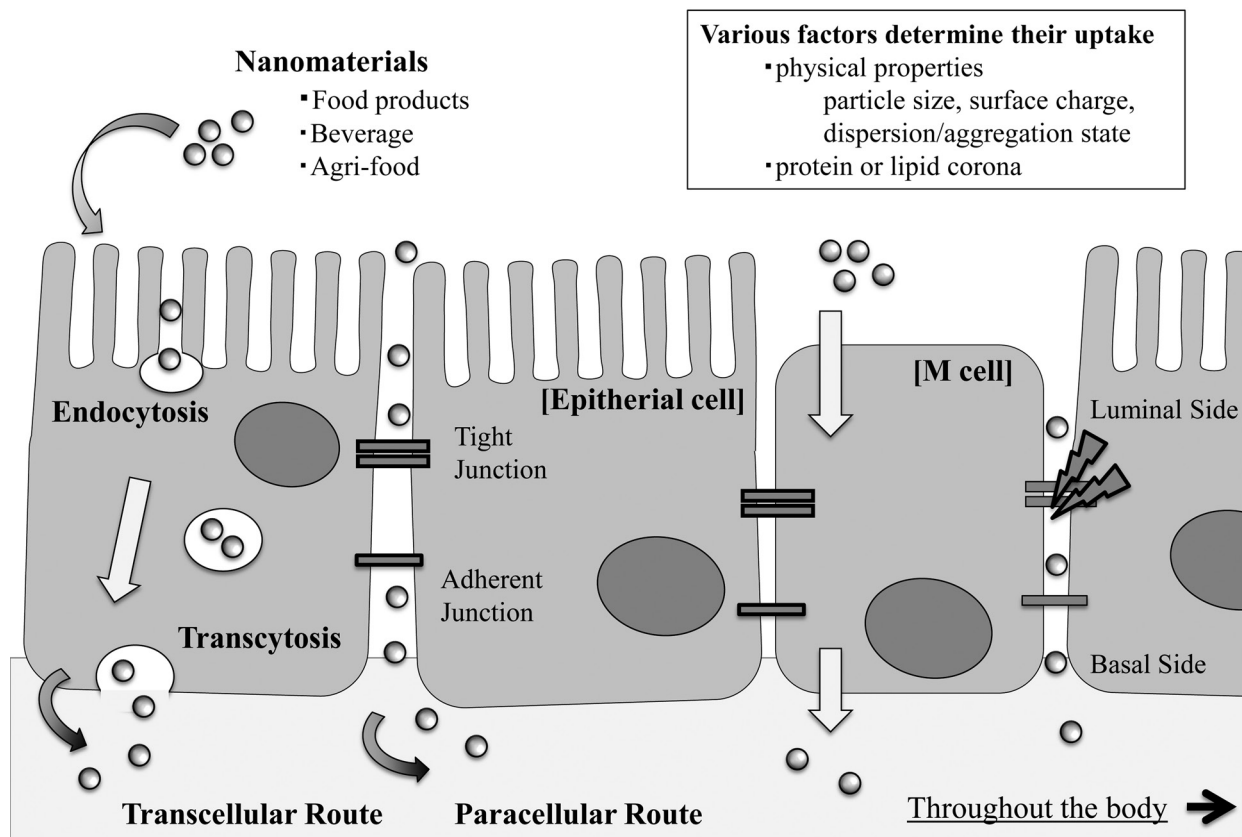

Fig. 1. The various absorption route of particles across intestinal barriers.

the immune system[29,30]. It is generally believed that nanoparticles in the intestine are taken up by enterocytes and goblet cells as well as by M-cells in the Peyer's patches and the isolated follicles of the intestinal-associated lymphoid tissue[31,32]. In fact, Brun et al reported that translocation of titanium dioxide nanoparticles through the regular ileum epithelium at Peyer's patches can induce epithelium impairment[33]. Therefore, it is reasonable to assume that nanoparticles can be absorbed through epithelial cells.

Cellular uptake of nanoparticles depends on their physical properties, such as particle size, surface charge, and dispersion or aggregation state[34,35]. To clarify a major role of surface properties in the extent of intestinal absorption of silica nanoparticles, we used an everted gut sac model where intestinal sacs from rats were incubated in solutions containing silica particles. Using inductively coupled plasma optical emission spectrometry, we found that silica nanoparticles modified with a carboxyl group or an amino group was more readily absorbed from the mucosal side to the serosal side of the sacs than were unmodified nanoparticles[36]. Schleh et al studied how the physical properties of gold nanoparticles affected the amount of the nanoparticles that passed through the intestinal membranes in adult female rats treated with gold nanoparticles of five different sizes (1.4–200 nm) and opposite surface charges. They revealed that absorption across the intestinal barrier decreased with increasing particle size and that negatively charged particles (2.8 nm) were more readily absorbed than positively charged particles (2.8 nm)[37]. Various factors have been suggested to affect intracellular transitivity of nanomaterials. The formation of a protein corona or lipid corona on the surface of nanomaterials may change their interaction with various receptors expressed on the surface of cells[38,39]. Specifically, the formation of a protein corona alters the physicochemical characteristics of nanoparticles, such as size and interfacial composition, which can in turn impart new biological characteristics to the nanomaterials and change their effects on cells exposed to them.

4. Types of Nanomaterials Used in Food and Evaluation of Their Safety

The major nanomaterials used in consumer products are silver, silica, and titanium dioxide. Among different product categories, silver nanomaterials are the most widely used, and particularly common in products in the food and beverage category and the home and garden category[40,41].

4.1. Applications and Safety of Silver Nanoparticles Used in the Food Industry

Owing to the antimicrobial activity of silver, silver nanoparticles are widely used in consumer products[14,42]. The mechanism of the antibacterial activity of silver nanoparticles has not been elucidated, but they may interact with the membranes of bacteria. The antibacterial activity of silver nanoparticles is likely due in part to Ag^+ ions readily formed on the surface of the nanoparticles through the reaction with oxygen, and the antibacterial activity of silver nanoparticles increases with decreasing particle size, which has been attributed to the increase in the surface area to mass ratio as particle size decreases[43].

Growing amounts of evidence suggest that orally ingested silver nanoparticles can cross the gastrointestinal barrier, be absorbed into the blood[44], and cause adverse health effects. Kim et al demonstrated that 56-nm silver nanoparticles can be found in rat blood, the liver, and other organs after 90 days of oral exposure at a dose of 30, 125, or 500 mg/kg bw/day. These investigators reported a no-observed-adverse-effect level (NOAEL) of 30 mg/kg bw/day and a lowest-observed-adverse-effect level (LOAEL) of 125 mg/kg bw/day[45]. It is reported that when rats were orally administered for 28 consecutive days with suspensions of uncoated silver nanoparticles with diameters of <20 nm or with suspensions of polyvinylpyrrolidone-coated nanoparticles with diameters of <15 nm, the nanoparticles were distributed in all the organs, but mainly in the liver[44,46]. Based on comparisons of the organ concentrations of silver between the nanoparticles-treated and silver nitrate-treated rats, they suggested that mainly Ag^+ ion, rather than silver nanoparticles, passed the intestines of rats[45]. In another study in rats, localization of silver in the liver was detected in several cell types, such as Kupffer cells and sinusoidal endothelium cells[45,47]. However, Van der Zande et al reported that in rats exposed to silver nanoparticles for 28 days, plasma levels of alanine aminotransferase and aspartate transaminase were relatively low compared with control values and that there were no significant differences between the silver-treated groups and the respective control groups. These investigators suggested that the silver nanoparticles induced no acute hepatotoxicity[44].

4.2. Applications and Safety of Silica Nanoparticles Used in the Food Industry

Amorphous silica nanoparticles are widely used in food products, for example, as thickeners, anticaking agents, and carriers for fragrances and flavors[48–50]. Nanosilica (E551) is registered as a food additive in the European Union; Dekkers et al analyzed food products including E551 and found that they contained silica nanoparticles at concentrations ranging from <0.1 to 6.9 mg/g product and with particle sizes ranging from 30 to 200 nm[51]. According to the Scientific Committee on Food of the European Food Safety Authority (EFSA), the estimated intake of silicon derived from silica used as an additive is 20–50 mg silicon per day in a 60 kg person[52].

Fu et al assessed the absorption, distribution, metabolism, excretion, and toxicity of 110 nm silica nanoparticles via various exposure routes in mice[53]. These investigators found that the nanoparticles through intramuscular or hypodermic injection passed through biological barriers such as the intestine. The particles were further transported to the liver, although the extent of absorption was low. Fu et al showed that orally administered silica nanoparticles were taken up into the intestinal tract. They also demonstrated that nanoparticles administered intravenously were present mainly in the liver and spleen and that they were excreted mainly in urine and feces[53].

Fruijtier-Polloth et al evaluated the toxic effects and safety of silica nanoparticles and concluded that they are as safe as conventional silica particles[48]. The toxicity of amorphous silica nanoparticles has been assessed both at the cellular level and in animal models. For example, on the basis of the results of a study using human gastric epithelial GES-1 cells and colorectal adenocarcinoma Caco-2 cells, Yang et al suggested that silica nanoparticles (200–450 nm) may arrest the cell cycle and inhibit cell growth but they do not induce apoptosis or necrosis[54]. We previously demonstrated that silicon was not detected in the liver or blood of mice after a 28-day daily oral exposure at a total dose of about 3.5 g silica particles/kg body weight, and that none of the particles tested showed toxic biological effects[36].

So et al studied the toxic effects of silica nano- and microparticles on mice at a total dose of 140 g silica particles/kg body weight[55]. These investigators found that the plasma level of alanine transaminase in mice treated with silica nanoparticles (30 nm) was higher than the level in mice treated with silica microparticles (30 μm). The amount of silicon in the livers of the mice treated with silica nanoparticles was lower than that in the livers of the mice treated with the silica microparticles. The authors concluded that the reactivity of silica nanoparticles in mouse liver was higher than that of silica microparticles[55].

In a sub-chronic toxicity study, Van der Zande et al found that oral exposure to silica nanoparticles at 2500 mg/kg body weight per day had biological effects on rat liver[56]. However, the authors pointed out that dose-response relationships should be assessed at doses that are more relevant to actual exposure levels. Although the safety and potential toxicity of silica nanoparticles need to be yet clarified toward a conclusive evaluation at present, an acceptable daily intake (ADI) of it should be specified and the guideline for preventing its excess intake over the estimated ADI needs to be considered.

4.3. Titanium Dioxide Nanoparticles in Food Products

Titanium dioxide nanoparticles are one of the top five types of nanomaterials used in consumer products such as cosmetics, as well as in food products, paints, and medicines[57]. Titanium dioxide nanoparticles occur in three main forms, i.e., anatase, rutile, and brookite[58], each of which has unique properties and uses. For example, the rutile form is used in pigments and sunscreens, and the anatase form is used in printing inks and photocatalysts[59]. Titanium dioxide has been approved by USFDA[60] and is widely used as a white pigment and food colorant[61]. The daily intake of titanium dioxide from food is estimated to be 5.4 mg/person /day (about 90 μg/kg bw/day) in the United Kingdom[62]. In Europe, food-grade titanium dioxide is designated as E171, and approximately 36% of the particles in E171 have diameters of <100 nm[63]. Candies, sweets, and chewing gums have the highest content of titanium (0.01–1 mg titanium per serving)[33]. By separating titanium particles from food products, Chen et al showed that more than the expected number of foods contained titanium dioxide nanoparticles with diameters of <200 nm[62]. The same authors also showed that the titanium dioxide in gum, more than 93% of which are nanoparticles, easily comes out of the gum during chewing and is ingested[62].

Many researchers have assessed the *in vivo* and *in vitro* toxicity of titanium dioxide nanoparticles[64]. For example, Warheit et al found that oral administration of titanium dioxide nanoparticles (140 nm diameter) in water to rats at 175, 550, 1750 or 5000 mg/kg bw/day resulted in no mortality or no clear body weight losses[65]. Wang et al showed that when mice were treated with titanium dioxide nanoparticles with diameters of 25 or 80 nm at a dose of 5000 mg/kg body weight, various biochemical parameters such as levels of alanine transaminase, aspartate transaminase, blood urea nitrogen, and lactase dehydrogenase were changed along with lesions on the liver and kidneys of female mice[66]. In addition,

Bu et al investigated the toxic effects of titanium dioxide nanoparticles administered orally to Wistar rats for 14 days by using conventional approaches and metabolomic analysis. These investigators found that at a dose of 1000 mg/kg body weight per day, the nanoparticles interfered with energy and amino acid metabolism and disrupted the gut microflora[67]. It is important to note that the doses of titanium dioxide used in these studies were approximately 2200–5500 times of the estimated human intake through food consumption. Cho et al found that even after titanium dioxide nanoparticles were orally administered to rats for 13 weeks (7 days/week), absorption of the nanoparticles was extremely low, and even at the highest dose (1041.5 mg/kg body weight per day), the amounts of nanoparticles in the sampled organs were not significantly higher than the amounts in the control group[68].

5. Conclusion

Although the history of nanomaterial use in the food sector is not long, recent rapid progress in the development of nanomaterials has prompted their safety concern worldwide. At a meeting of experts of the Food and Agriculture Organization of the United Nations and the World Health Organization, the tremendous benefits of nanomaterials in the food sector were acknowledged, but it was suggested that a taxonomy should be developed to help with risk management and that clear and international definitions should be devised for the application of nanomaterials[69]. In the United States, the Food and Drug Administration Nanotechnology Task Force was established in 2006 to devise regulatory policies to manage the use of nanomaterials in the food sector[70]. Although all of these organizations agree in principle that nanomaterials will bring significant benefits to the food sector, they also agree that the current safety information about nanomaterials, as well as technologies and data for risk analysis and assessment, is insufficient and that this issue must be addressed.

Risk assessment guidelines for the use of nanomaterials in the food sector were announced by EFSA in 2011[71] and by USFDA in 2012[72]. These agencies stressed the importance not only of hazard analyses but also of analysis of the associations between intestinal absorption and characteristics such as particle size and surface properties. Since then, little progress has been made in terms of safety assessments of nanomaterials in the food sector worldwide. Available information about safety after oral intake of nanomaterials is minimal, and absorption, distribution, metabolism, and excretion information that is essential for risk analysis is yet poorly understood. Moreover, although we are beginning to understand how characteristics of nanomaterials such as particle size, shape, surface properties, and dispersion and aggregation state might influence the biological effects, studies on the safety of nanomaterials have not reached the stage at which the results can be used to control quality, develop guidelines for nanomaterials, and regulate security of nanomaterials. Therefore, quantitative analyses of these characteristics of nanomaterials are required, along with assessments of the relationships between these characteristics and the effectiveness, tissue distribution, and safety. Such researches are expected to provide the scientific basis for the risk assessments that are necessary for the development and safe use of nanomaterials in the food sector.

Acknowledgement

References

1. Scholars WWICf Consumer products inventor, food and beverage category. http://www.nanotechproject.org/cpi/browse/categories/food-and-beverage/?x. Accessed on January 24, 2015.

2. Tachikawa M. Uncertainty of, and stakeholder response to, emerging technologies: Food nanotechnology in japan. Ethics Sci Environ Polit. 2012; **12**: 113–122.

3. Cockburn A, Bradford R, Buck N, et al. Approaches to the safety assessment of engineered nanomaterials (ENM) in food. Food Chem Toxicol. 2012; **50**(6): 2224–2242. [Medline]

4. Martirosyan A, Schneider YJ. Engineered nanomaterials in food: implications for food safety and consumer health. Int J Environ Res Public Health. 2014; **11**(6): 5720–5750. [Medline]

5. Wang H, Du LJ, Song ZM, Chen XX. Progress in the characterization and safety evaluation of engineered inorganic nanomaterials in food. Nanomedicine (Lond). 2013; **8**(12): 2007–2025. [Medline]

6. Mukhopadhyay SS. Nanotechnology in agriculture: prospects and constraints. Nanotechnol Sci Appl. 2014; **7**: 63–71. [Medline]

7. Sekhon BS. Nanotechnology in agri-food production: an overview. Nanotechnol Sci Appl. 2014; **7**: 31–53. [Medline]

8. Kuzma J. Moving forward responsibly: Oversight for the nanotechnology-biology interface. Journal of Nanoparticle Research. 2007; **9**: 165–182.

9. Rai M, Ingle A. Role of nanotechnology in agriculture with special reference to management of insect pests. Appl Microbiol Biotechnol. 2012; **94**(2): 287–293. [Medline]

10. Luykx DM, Peters RJ, van Ruth SM, Bouwmeester H. A review of analytical methods for the identification and characterization of nano delivery systems in food. J Agric Food Chem. 2008; **56**(18): 8231–8247. [Medline]

11. Sekhon BS. Food nanotechnology - an overview. Nanotechnol Sci Appl. 2010; **3**: 1–15. [Medline]

12. Huang Q, Yu H, Ru Q. Bioavailability and delivery of nutraceuticals using nanotechnology. J Food Sci. 2010; **75**(1): R50–R57. [Medline]

13. Qi L, Xu Z, Jiang X, Hu C, Zou X. Preparation and antibacterial activity of chitosan nanoparticles. Carbohydr Res. 2004; **339**(16): 2693–2700. [Medline]

14. Morones JR, Elechiguerra JL, Camacho A, et al. The bactericidal effect of silver nanoparticles. Nanotechnology. 2005; **16**(10): 2346–2353. [Medline]

15. Cho M, Chung H, Choi W, Yoon J. Different inactivation behaviors of MS-2 Phage and *Escherichia coli* in TiO_2 photocatalytic disinfection. Appl Environ Microbiol. 2005; **71**(1): 270–275. [Medline]

16. Chaudhry Q, Scotter M, Blackburn J, et al. Applications and implications of nanotechnologies for the food sector. Food Addit Contam Part A Chem Anal Control Expo Risk Assess. 2008; **25**(3): 241–258. [Medline]

17. Xu B, Zheng Q, Song YH, Shangguan Y. Calculating barrier properties of polymer/clay nanocomposites: Effects of clay layers. Polymer. 2006; **47**: 2904–2910.

18. R Kotsilkova VP Y Pelovski. Thermal analysis of polymer–silicate nanocomposites. Journal of Thermal Analysis and Calorimetry. 2001; **64**: 591–598.

19. Kuang H, Chen W, Yan W, et al. Crown ether assembly of gold nanoparticles: melamine sensor. Biosens Bioelectron. 2011; **26**(5): 2032–2037. [Medline]

20. Guan M, Zhu QL, Liu Y, et al. Uptake and transport of a novel anticancer drug-delivery system: lactosyl-norcantharidin-associated N-trimethyl chitosan nanoparticles across intestinal Caco-2 cell monolayers. Int J Nanomedicine. 2012; **7**: 1921–1930. [Medline]

21. Saremi S, Dinarvand R, Kebriaeezadeh A, Ostad SN, Atyabi F. Enhanced oral delivery of docetaxel using thiolated chitosan nanoparticles: Preparation, in vitro and in vivo studies. Biomed Res Int. 2013;2013:150478.

22. He B, Lin P, Jia Z, et al. The transport mechanisms of polymer nanoparticles in Caco-2 epithelial cells. Biomaterials. 2013; **34**(25): 6082–6098. [Medline]

23. Moghimi SM, Hunter AC, Murray JC. Nanomedicine: current status and future prospects. FASEB J. 2005; **19**(3): 311–330. [Medline]

24. Tang VW, Goodenough DA. Paracellular ion channel at the tight junction. Biophys J. 2003; **84**(3): 1660–1673. [Medline]

25. Ruenraroengsak P, Cook JM, Florence AT. Nanosystem drug targeting: Facing up to complex realities. J Control Release. 2010; **141**(3): 265–276. [Medline]

26. Amidi M, Mastrobattista E, Jiskoot W, Hennink WE. Chitosan-based delivery systems for protein therapeutics and antigens. Adv Drug Deliv Rev. 2010; **62**(1): 59–82. [Medline]

27. Sonaje K, Lin KJ, Tseng MT, et al. Effects of chitosan-nanoparticle-mediated tight junction opening on the oral absorption of endotoxins. Biomaterials. 2011; **32**(33): 8712–8721. [Medline]

28. Powell JJ, Faria N, Thomas-McKay E, Pele LC. Origin and fate of dietary nanoparticles and microparticles in the gastrointestinal tract. J Autoimmun. 2010; **34**(3): J226–J233. [Medline]

29. Gebert A, Rothkötter HJ, Pabst R. M cells in Peyer's patches of the intestine. Int Rev Cytol. 1996; **167**: 91–159. [Medline]

30. Beier R, Gebert A. Kinetics of particle uptake in the domes of Peyer's patches. Am J Physiol. 1998; **275**(1 Pt 1): G130–G137. [Medline]

31. Florence AT. The oral absorption of micro- and nanoparticulates: neither exceptional nor unusual. Pharm Res. 1997; **14**(3): 259–266. [Medline]

32. Seifert J, Sass W. Intestinal absorption of macromolecules and small particles. Dig Dis. 1990; **8**(3): 169–178. [Medline]

33. Brun E, Barreau F, Veronesi G, et al. Titanium dioxide nanoparticle impact and translocation through ex vivo, in vivo and in vitro gut epithelia. Part Fibre Toxicol. 2014; **11**: 13. [Medline]

34. Geiser M, Rothen-Rutishauser B, Kapp N, et al. Ultrafine particles cross cellular membranes by nonphagocytic mechanisms in lungs and in cultured cells. Environ Health Perspect. 2005; **113**(11): 1555–1560. [Medline]

35. He C, Hu Y, Yin L, Tang C, Yin C. Effects of particle size and surface charge on cellular uptake and biodistribution of polymeric nanoparticles. Biomaterials. 2010; **31**(13): 3657–3666. [Medline]

36. Yoshida T, Yoshioka Y, Takahashi H, et al. Intestinal absorption and biological effects of orally administered amorphous silica particles. Nanoscale Res Lett. 2014; **9**(1): 532. [Medline]

37. Schleh C, Semmler-Behnke M, Lipka J, et al. Size and surface charge of gold nanoparticles determine absorption across intestinal barriers and accumulation in secondary target organs after oral administration. Nanotoxicology. 2012; **6**(1): 36–46. [Medline]

38. Monopoli MP, Aberg C, Salvati A, Dawson KA. Biomolecular coronas provide the biological identity of nanosized materials. Nat Nanotechnol. 2012; **7**(12): 779–786. [Medline]

39. Shannahan JH, Podila R, Aldossari AA, et al. Formation of a protein corona on silver nanoparticles mediates cellular toxicity via scavenger receptors. Toxicol Sci. 2015; **143**(1): 136–146. [Medline]

40. Hansen SF, Michelson ES, Kamper A, Borling P, Stuer-Lauridsen F, Baun A. Categorization framework to aid exposure assessment of nanomaterials in consumer products. Ecotoxicology. 2008; **17**(5): 438–447. [Medline]

41. Fröhlich E, Roblegg E. Models for oral uptake of nanoparticles in consumer products. Toxicology. 2012; **291**(1-3): 10–17. [Medline]

42. Ji JH, Jung JH, Kim SS, et al. Twenty-eight-day inhalation toxicity study of silver nanoparticles in Sprague-Dawley rats. Inhal Toxicol. 2007; **19**(10): 857–871. [Medline]

43. Lok CN, Ho CM, Chen R, et al. Silver nanoparticles: partial oxidation and antibacterial activities. J Biol Inorg Chem. 2007; **12**(4): 527–534. [Medline]

44. van der Zande M, Vandebriel RJ, Van Doren E, et al. Distribution, elimination, and toxicity of silver nanoparticles and silver ions in rats after 28-day oral exposure. ACS Nano. 2012; **6**(8): 7427–7442. [Medline]

45. Kim YS, Song MY, Park JD, et al. Subchronic oral toxicity of silver nanoparticles. Part Fibre Toxicol. 2010; **7**: 20. [Medline]

46. Loeschner K, Hadrup N, Qvortrup K, et al. Distribution of silver in rats following 28 days of repeated oral exposure to silver nanoparticles or silver acetate. Part Fibre Toxicol. 2011; **8**: 18. [Medline]

47. Danscher G. Light and electron microscopic localization of silver in biological tissue. Histochemistry. 1981; **71**(2): 177–186. [Medline]

48. Fruijtier-Pölloth C. The toxicological mode of action and the safety of synthetic amorphous silica-a nanostructured material. Toxicology. 2012; **294**(2-3): 61–79. [Medline]

49. Peters R, Kramer E, Oomen AG, et al. Presence of nano-sized silica during *in vitro* digestion of foods containing silica as a food additive. ACS Nano. 2012; **6**(3): 2441–2451. [Medline]

50. Dekkers S, Bouwmeester H, Bos PM, Peters RJ, Rietveld AG, Oomen AG. Knowledge gaps in risk assessment of nanosilica in food: evaluation of the dissolution and toxicity of different forms of silica. Nanotoxicology. 2013; **7**(4): 367–377. [Medline]

51. Dekkers S, Krystek P, Peters RJ, et al. Presence and risks of nanosilica in food products. Nanotoxicology. 2011; **5**(3): 393–405. [Medline]

52. European Food Safety Authority. Opinion of the scientific panel on dietetic products, nutrition and allergies on a request from the commission related to the tolerable upper intake level of silicon EFSA Journal. 2004;60:1–11. Available at: http://www.efsa.europa.eu/en/scdocs/doc/60.pdf.

53. Fu C, Liu T, Li L, Liu H, Chen D, Tang F. The absorption, distribution, excretion and toxicity of mesoporous silica nanoparticles in mice following different exposure routes. Biomaterials. 2013; **34**(10): 2565–2575. [Medline]

54. Yang YX, Song ZM, Cheng B, et al. Evaluation of the toxicity of food additive silica nanoparticles on gastrointestinal cells. J Appl Toxicol. 2014; **34**(4): 424–435. [Medline]

55. So SJ, Jang IS, Han CS. Effect of micro/nano silica particle feeding for mice. J Nanosci Nanotechnol. 2008; **8**(10): 5367–5371. [Medline]

56. van der Zande M, Vandebriel RJ, Groot MJ, et al. Sub-chronic toxicity study in rats orally exposed to nanostructured silica. Part Fibre Toxicol. 2014; **11**: 8. [Medline]

57. Shukla RK, Sharma V, Pandey AK, Singh S, Sultana S, Dhawan A. ROS-mediated genotoxicity induced by titanium dioxide nanoparticles in human epidermal cells. Toxicol In Vitro. 2011; **25**(1): 231–241. [Medline]

58. Macwan DPPN, Shalini C. A review on nano-TiO$_2$ sol–gel type syntheses and its applications. Journal of Materials Science. 2011; **46**: 3669–3686.

59. Morishige T, Yoshioka Y, Tanabe A, et al. Titanium dioxide induces different levels of IL-1β production dependent on its particle characteristics through caspase-1 activation mediated by reactive oxygen species and cathepsin B. Biochem Biophys Res Commun. 2010; **392**(2): 160–165. [Medline]

60. Banerjee K, Thiagarajan P. A review of titanium di oxide nanoparticles − synthesis, applications and toxicity concerns. Nanoscience & Nanotechnology-Asia. 2014; **4**: 132–143.

61. Buettner KM, Valentine AM. Bioinorganic chemistry of titanium. Chem Rev. 2012; **112**(3): 1863–1881. [Medline]

62. Chen XX, Cheng B, Yang YX, et al. Characterization and preliminary toxicity assay of nano-titanium dioxide additive in sugar-coated chewing gum. Small. 2013; **9**(9-10): 1765–1774. [Medline]

63. Weir A, Westerhoff P, Fabricius L, Hristovski K, von Goetz N. Titanium dioxide nanoparticles in food and personal care products. Environ Sci Technol. 2012; **46**(4): 2242–2250. [Medline]

64. Shi H, Magaye R, Castranova V, Zhao J. Titanium dioxide nanoparticles: a review of current toxicological data. Part Fibre Toxicol. 2013; **10**: 15. [Medline]

65. Warheit DB, Hoke RA, Finlay C, Donner EM, Reed KL, Sayes CM. Development of a base set of toxicity tests using ultrafine TiO$_2$ particles as a component of nanoparticle risk management. Toxicol Lett. 2007; **171**(3): 99–110. [Medline]

66. Wang J, Zhou G, Chen C, et al. Acute toxicity and biodistribution of different sized titanium dioxide particles in mice after oral administration. Toxicol Lett. 2007; **168**(2): 176–185. [Medline]

67. Bu Q, Yan G, Deng P, et al. NMR-based metabonomic study of the sub-acute toxicity of titanium dioxide nanoparticles in rats after oral administration. Nanotechnology. 2010; **21**(12): 125105. [Medline]

68. Cho WS, Kang BC, Lee JK, Jeong J, Che JH, Seok SH. Comparative absorption, distribution, and excretion of titanium dioxide and zinc oxide nanoparticles after repeated oral administration. Part Fibre Toxicol. 2013; **10**: 9. [Medline]

69. Food and Agriculture Organization of the United Nations and World Health Organization. FAO/WHO expert meeting on the application of nanotechnologies in the food and agriculture sectors: Potential food safety implications, meeting report. 2010. Available at: http://www.fao.org/docrep/012/i1434e/i1434e00.pdf.

70. FDA R, MD. Nanotechnology: A report of the US food and drug administration nanotechnology task force. 2006.

71. European Food Safety Authority. Guidance on the risk assessment of the application of nanoscience and nanotechnologies in the food and feed chain. EFSA Journal. 2011;9:2140. Available at: http://www.efsa.europa.eu/en/efsajournal/doc/2140.pdf.

72. Food and Drug Administration. Guidance for industry: Assessing the effects of significant manufacturing process changes, including emerging technologies, on the safety and regulatory status of food ingredients and food contact substances, including food ingredients that are color additives; availability. Notices. Federal Register. 2014; **79**: 36533–36534.