

FATE, TRANSPORT AND TOXICITY OF NANOMATERIALS IN DRINKING WATER

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ABSTRACT

The properties and fate of nanomaterials in water affects their potential environmental impacts and risk for human exposure. In this study, the physicochemical characteristics of six commercial metal oxide nanoparticles, a lab-synthesized hematite nanoparticle and a water-soluble CdTe quantum dots (QDs), their removal from water by conventional drinking water treatment processes and their toxicity on cells were examined. These nanomaterials exhibited different stability in water and conventional water treatment could not remove them completely. Toxicity studies showed that TiO₂ nanomaterials could flatten down the microvilli of cells, decrease Trans-epithelial Electrical Resistance (TEER) and pass through the epithelial cells. These results imply that nanomaterials in drinking water may pose a potential exposure risk for human.

Keywords: aggregate, stability, flocculation, removal, toxicity

INTRODUCTION

Nanomaterials, darlings of the science and technology in the 21st century, are extensively being applied in commercial products, as a result of their novel and desirable properties. The current market for nanomaterials is increasing rapid and it is estimated that annual production of nanomaterials will be the order of millions of tons by 2010¹. Accordingly, it can be predictable that significant quantity of nanomaterials will show up in our daily life and be inevitably introduced into environment in the near future. As a category of emerging materials in environment, nanomaterials are receiving a lot of attention with their adverse effects on environment and human health. Recent research indicates that nanomaterials in environment can enter into human body by inhalation, ingestion and the skin²⁻³ and exhibit a certain degree of toxicity⁴⁻⁵. Thus, nanomaterials may pose a significant problem for the human health.

Drinking water consumption is considered to be an important route for nanomaterials to enter human body. Although EPA enacted the regulations for particles in drinking water, current efforts for removal of particles from water may have little influence on nanomaterials. The assessment on potential risk of human exposure to

nanomaterials in water required the fundamental understanding on their fate and transport in drinking water and their toxicity for human. However, the fate and toxicity of nanomaterials still has been poorly studied. Accordingly, this research examines the following three issues: 1) characteristics and stability of nanomaterials in the water environment; 2) removal of nanomaterials during conventional water treatment processes; and 3) the toxicity of nanomaterials on cells.

MATERIALS AND METHODS

Six commercial metal oxide nanoparticles, including two types of TiO₂, Fe₂O₃, ZnO, NiO and silica (Sigma-Aldrich), a lab-synthesized nanoparticle (Hematite) and a commercial quantum dots (American Dye Source Inc.) were used in this research (Table 1). Hematite (α -Fe₂O₃) nanoparticles were synthesized and stored in 4 mM HCl, using the method that was describe by Penners 1986⁶. Quantum dots were composed by CdTe core and sodium thioglycolate (HS-CH₂-COONa) capping ligands.

Table 1. Summary of nanomaterial properties

Nano-material	Diameter (nm)		Number concentration (> 3 μ m)	pH _Z _{PC}
	Vendor reported	DLS		
TiO ₂ (powder)	15	530 \pm 30	9300 \pm 300	5.2
TiO ₂ (5wt% suspension)	40	200 \pm 10	9000 \pm 200	5.2
Fe ₂ O ₃ (powder)	5~25	200 \pm 10	3500 \pm 200	6.5
ZnO (powder)	50~70	320 \pm 20	4400 \pm 200	9.2
NiO (powder)	10~20	750 \pm 30	45000 \pm 3000	9.1
Silica (powder)	10	740 \pm 40	20000 \pm 2000	1.8
Hematite (1wt% suspension)	80~90 based on SEM	85 \pm 3	NA	8.4
CdTe QDs (0.25 Cd% suspension)	3.5~4.5	80 \pm 20	NA	NA

Preparation of nanomaterial suspensions

1 g/L nanomaterial stock suspensions were prepared by suspending 1 g of dry nanopowder or adding a certain volume of the as-received nanomaterial suspension to 1 L of nanopure water (conductivity <5 $\mu\text{S}/\text{cm}$, $\text{pH}=5.6\pm 0.2$). The stock suspension was then sonicated for 15 minutes at 20 KHz at an intensity of 200 W/L (Model 2000U, Ultrasonic Power Corp.) and then stored at room temperature (25°C) for no longer than 2 days. 10 mg/L nanomaterial suspensions were prepared by diluting the stock suspensions into nanopure water in all experiments, except where noted.

Characterization of nanomaterials

Particle sizes of nanomaterials in water were examined by dynamic light scattering (DLS, 90 Plus Particle Size Analyzer, Brookhaven Instrument Corp.), micro-flow imaging (MFI, DFA 4100, Brightwell Technologies Inc.), scanning electron microscopy (SEM, Philips XL30, FEI) and transmission electron microscopy (TEM, JEM-2000FX, JEOL). DLS can only measure particles with sizes ranging from 2 nm to 3 μm and the average diameter is weighted by light scattering intensity. The particles ranging from 3 to 400 μm and corresponding number concentrations were measured by MFI. SEM and TEM samples were prepared by drying several drops of the 10 mg/L nanomaterial suspension on the stage and a 300-mesh copper grid, respectively.

Zeta Potentials of nanomaterials were determined from electrophoretic mobility in suspension with 0.01 M KNO_3 as background electrolyte, using ZetaPlus Analyzer (Brookhaven Instruments Corp.).

Disaggregation of nanomaterials

Ultrasonic and dispersants were used to disperse commercial TiO_2 nanopowders in water. Sonication was conducted at a frequency of 20 KHz and input energy at 200 W/L for 10 to 30 minutes. Seven dispersants included 1 to 10 mM sodium dodecyl sulfonate (SDS), 1 to 10 g/L sodium hexametaphosphate ($(\text{NaPO}_3)_6$), or 10% by volume of acetone, butanonone, methanol, isopropanol, and ethylene glycol. Suspension without any dispersant was prepared using the identical procedure and the pH of all suspensions was adjusted to ~ 7.0 . In addition, the disaggregation of TiO_2 under acidic condition ($\text{pH} = 2$) was examined.

Wet flocculated hematite and dry hematite powders were employed to evaluate disaggregation of lab-synthesized hematite nanoparticles. Wet flocculated hematite nanoparticles were prepared by adding 0.1 M MgCl_2 into hematite suspension. Dry hematite powders were obtained by drying the hematite suspension using a freeze dryer with an operating temperature of -40°C and pressure at 1.03 mbar (Free Zone 7753522, Labconco Corp.). These two types of hematite nanoparticles were

then stored at room temperature for a certain period of time. Then they were re-suspended in 4 mM HCl and sonicated for 10 minutes. Before being re-suspended, wet flocculated hematite was separated from the suspensions containing MgCl_2 by centrifugation.

Stability and flocculation of nanomaterials

10 mg/L nanomaterial suspensions were prepared by adding stock suspension into nanopure water or tap water (conductivity = 2000~2500 $\mu\text{S}/\text{cm}$, $\text{pH} = 8.1\pm 0.2$). The suspension was mixed thoroughly for 30 seconds and then the change of particle size was measured over time. The number concentration of particles larger than 3 μm was measured every two hours.

Flocculation of 10 mg/L metal oxide nanoparticles in 0.01 to 0.1 M KCl solutions and 0.1 M MgCl_2 solution was examined. The pH of all suspensions was adjusted to ~ 8.0 . Zeta potentials and particle sizes of nanoparticles in suspensions were monitored.

Flocculation of 0.25 g Cd/L quantum dots in the presence of mono-valent (KCl), divalent (MgCl_2 and CaCl_2) and trivalent ($\text{Al}_2(\text{SO}_4)_3$) electrolytes was evaluated using the above identical procedures at pH 8 and 5. K^+ concentration ranged from 10 to 150 meq/L. Divalent and trivalent cations were within a concentration range of 0 to 10 meq/L.

Removal of nanomaterials in water treatment

Nanomaterials removal in nanopure water buffered with 10 mM NaHCO_3 ($\text{pH} 8.2\pm 0.2$) and tap water was evaluated using jar tests. 0.1 M MgCl_2 and alum ($\text{Al}_2(\text{SO}_4)_3 \cdot 16\text{H}_2\text{O}$) with different dosage were added into 2-L jars containing 10 mg/L nanomaterials suspensions. Jar tests were conducted as the following three steps: (1) rapid mixing for 1 minute at 100 rpm, ($G = 99.5 \text{ s}^{-1}$), (2) slow mixing for 30 minutes at 30 rpm ($G = 16.3 \text{ s}^{-1}$) and (3) settling for 1 hour. After sedimentation, TiO_2 nanoparticle supernatants were filtered using a 0.45 μm filter. The concentrations of nanomaterials were evaluated by measuring mass concentrations of metals by the digestion/GFAA method.

Toxicity of nanomaterials

Permeability of nanomaterials in the lumen of the digestive tract was evaluated by measuring TEER of Caco-2 cell line (human intestinal cells) grown in Dulbecco's Modified Eagle's Medium (DMEM). The cell line was cultured for approximately 7 days on collagen-treated permeable membrane inserts. Once the cells were grown to form a monolayer epithelial barrier, nanomaterials of 1 to 1000 mg/L was added onto the cells for further culture. Then, TEERs were measured at fixed time intervals. SEM images of Caco-2 cell line were taken to examine the influence of nanomaterials on cells.

RESULTS AND DISCUSSIONS

Characterization of nanomaterials

DLS measurements showed that all commercial metal oxide nanoparticles had bimodal size distributions in water and their average diameters were 200 to 800 nm, which was much larger than vendor reported values (Table 1). TEM and SEM images indicated that these commercial nanoparticles appeared to be aggregates, composed of primary nanoparticles less than 100 nm in water. In addition, MFI analysis showed that aggregates with sizes larger than 3 μm were more than 3000 per mL in 10 mg/L suspensions (Table 1). In contrast, lab-synthesized hematite had an average diameter of 85 nm and presented as mono-dispersed primary nanoparticles in water, which was demonstrated by DLS measurement and SEM images. Particle size distribution measured by DLS indicated that commercial CdTe QDs also could present as primary nanoparticles with sizes of 3.5 to 5 nm in water due to the presence of hydrophilic capping ligands.

Disaggregation of nanomaterials

As received, TiO_2 powders were aggregates larger than 200 nm. Before sonication, the average diameter of aggregated TiO_2 in water was above 1000 nm. After 10 minutes of sonication, the average diameter of TiO_2 decreased to approximately 500 to 550 nm. However, as the sonication time increased from 10 minutes to 30 minutes, the average diameters of TiO_2 nanoparticles still remained within this range. Therefore, sonication only partly disaggregated TiO_2 nanoparticles. Similar results were shown for all other commercial metal oxide nanoparticles. With $(\text{NaPO}_3)_6$ or SDS addition, although negative zeta potential of TiO_2 aggregates increased, its diameter did not change. TiO_2 nanoparticles all had the average diameters of 500 nm or larger, regardless of which dispersant was used. It implied that none of dispersants could break up these aggregates.

In contrast with the commercial nanomaterials, lab-synthesized hematite aggregates freshly produced by adding 0.1 M MgCl_2 could be disaggregated to individual nanoparticles completely in 4 mM HCl using sonication. However, if hematite nanoparticles were stored in wet flocculated state or as dry powders for more than one month, they formed irreversible aggregates that could not be disaggregated by sonication and average diameters were more than 100 nm. Particle size distribution analysis showed that more than 10% hematite of total volume could not be dispersed as primary nanoparticles. Therefore, irreversible aggregates of commercial metal oxide nanoparticles might originate from the storage.

Stability of nanomaterials in water

All studied nanomaterials, with the exception of silica, flocculated in tap water and were stable in nanopure water. Silica is the only nanoparticle to remain stable in both

waters, which may be related to its low Hamaker constant and high hydrophilicity. Commercial CdTe QDs exhibited fast flocculation in tap water, which may attribute to the presence of divalent cations.

Flocculation of nanomaterials with electrolyte

Low concentration of electrolyte (~ 0.03 M KCl) resulted in flocculation of metal oxide nanoparticles, except silica. With 0.1 M MgCl_2 , all metal oxide nanoparticles flocculated rapidly and their zeta potentials were within the range of -5 mV to +5 mV. However, QDs behaved quite different from metal oxide nanoparticles. It was observed that capping ligands stabilized QDs in high concentration KCl solution (0.1 M). However, a low concentration of divalent (Mg^{2+} & Ca^{2+}) and trivalent (Al^{3+}) cations reduced negative zeta potentials of QDs and led to their aggregation by forming complex with capping ligands. Because dominant species of hydrolytic Al^{3+} changed with pH, aggregation tendencies of QDs with Al^{3+} were different under basic and acidic conditions.

Removal of nanomaterials

Removal of nanomaterials by coagulation, flocculation and sedimentation processes was relatively difficult. At alum dosage up to 60 mg/L, more than 20% commercial metal oxide and lab-synthesized hematite nanoparticles by total mass still remained in settled water. Moreover, metal oxide nanoparticles in tap water had lower removal efficiencies than those in buffered nanopure water.

For CdTe QDs, as alum dosage was less than 100 mg/L, more than 80% of total mass could not be removed by sedimentation in nanopure water. Removal of 90% QDs required very high alum dosage (150 mg/L). However, in tap water, 80% or more QDs were removed from water even in the absence of alum because divalent cations in tap water destabilized them. Following sedimentation, 0.45 μm filtration was found to enhance removal efficiencies of all nanomaterials. However, there were still 5% to 10% nanomaterials remained in the finished water.

Toxicity of nanomaterials

Toxicity studies of nanomaterials on cells showed that TiO_2 nanomaterials reduce the length and number of the microvilli of cells and after chronic exposure, led to 25% decrease in Trans-epithelial Electrical Resistance (TEER) at very high concentration (1000 mg/L). In addition, 1% to 10% nanomaterials were found to pass through the epithelial cells.

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