Total dissolution and digestion methods for engineered metal nanoparticles

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Abstract

The application of engineered nanoparticles (ENPs) may lead to an increased occurrence in the environment. To assess the risk of metal NPs and to understand their behavior in the environment in different matrices ecotoxicological tests are required. For clear-cut conclusions it is often inevitable to quantify the total amount of added ENPs to the test systems. To achieve this task we developed different approaches for total dissolution and digestion of an assortment of metal NPs in various matrices. In this contribution the methods are described and performance characteristics are presented.

Introduction

Metal-nanoparticles are widely used in our daily life as well as in industry for example in textiles, cleaning products, cosmetics, and coatings [1]. Therefore, these engineered nanoparticles (ENPs) can reach the environment and may be distributed in different compartments of terrestrial and aqueous ecosystems. To assess the properties of ENPs with regard to their environmental relevance it is inevitable to develop a set of dedicated ecotoxicological tests and research methods [2]. For these procedures validated accompanying analytical methods are necessary. Therefore, different methods for total digestion and dissolution of nano-Ag, nano-TiO_2, and nano-Au in different matrices followed by quantification were developed and validated. Our set and fulfilled requirements for recoveries were 100 ± 25 %. These limits can only be reached by thoroughly homogenization of the sample material and complete dissolution or digestion, respectively. Table 1 compiles nanoparticles and matrices for which digestion/dissolution methods were developed and applied.

	nano-TiO_2	nano-Ag	nano-Au
aqueous samples	X	X	X
soil / sediment	X	X	X
WWTP sludge	-	X	-
earthworms	X	X	-

Table 1: Overview on developed dissolution and digestion methods for metal nanoparticles in different matrices. WWTP – waste water treatment plant.

Materials and methods

nano-Ag (material applied: NM-300K)

Digestion of nano-Ag in aqueous samples

Samples were thoroughly vortexed and 1 mL of this mixture was transferred into quartz digestion vessels. 2 mL of conc. nitric acid and 4 mL of water from an Ultra-Pure water preparation system were added, followed by digestion using an Ultra Clave II microwave digestion system (initial pressure 40 bar), heated up to 220°C in 25 min and remained at this temperature for additional 30 min. Thereafter, the digested samples were poured into volumetric flasks and filled up with ultra-pure water.

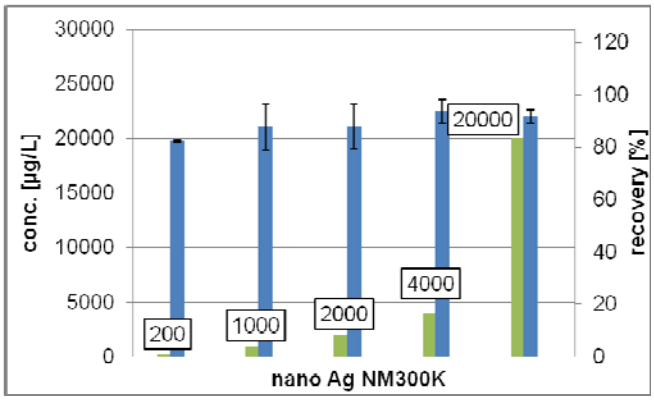

Figure 1: Aqueous medium spiked with known amounts of nano-Ag NM300K (left scale; green bars; nominal concentration values are printed in the diagram) and recoveries after digestion (right scale).

Digestion of nano-Ag in samples from sludge, sediment and soil. Digestion of nano-Ag was performed following ISO11466 using *aqua regia* [3]. After cooling, the resulting mixture was filtered (syringe filter 0.2 µm, supor membrane) and carefully filled up to an exact volume. Depending on the expected amount of nano-Ag and higher chloride concentrations in the sample more *aqua regia* or less sample weight for extraction and additional hydrochloric acid (formation of soluble $AgCl_2^-$) might help for complete dissolution.

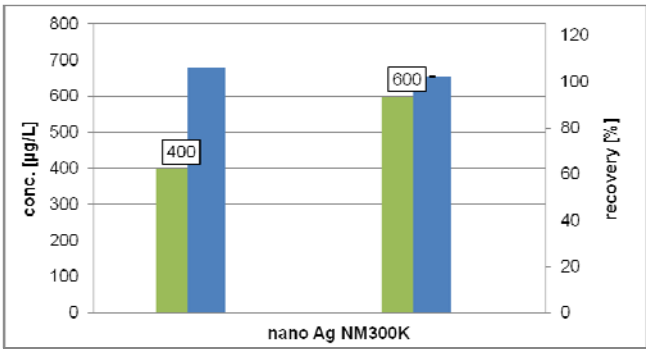

Figure 2: Spiked sludge suspension from WWTP with different concentrations of nano-Ag NM300K (left scale; green bars; nominal concentration values are printed in the diagram) and their recoveries after digestion (right scale).

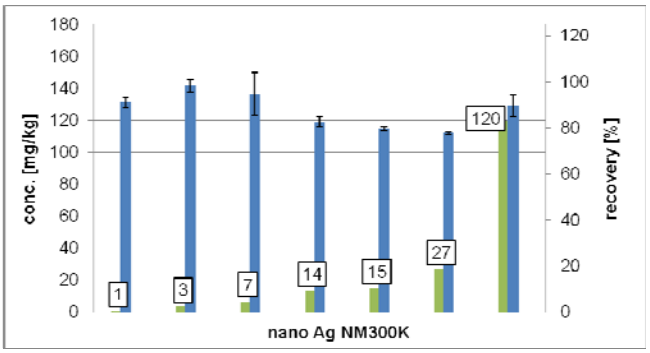

Figure 3: Sediment/soil spiked with known amounts of nano-Ag NM300K (left scale; green bars; nominal concentration values are printed in the diagram) and recoveries after digestion (right scale).

Digestion of nano-Ag in earthworms (*Lumbriculus terrestris*)
Cryogenic homogenisation [4] was necessary for a complete digestion of worms. Therefore samples were homogenised using pistils and a mortar (everything cooled in liquid nitrogen). The resulting frozen powder samples were lyophilized until constant weights were reached. Approx. 200 mg of the resulting material were digested in the microwave digestion system after adding 5 mL of concentrated nitric acid. The further procedure was as described above for the digestion of nano-Ag in aqueous samples.

Analytical quantification of Ag
Nano-Ag reference materials with certified amounts of Ag were not commercially available at the time of the development of these methods. Therefore, to validate the methods, chosen matrices were spiked with known amounts of nano-Ag, digested, measured and their recoveries were determined (Fig. 1-3). All final solutions were measured with matrix adjusted calibrations by ICP-OES (Thermo Scientific IRIS Intrepid II) or ICP-MS (Agilent 7500ce or 7700) for their amount of silver. Commercially available Ag ICP standards were applied for the calibration of the instruments.

nano-TiO_2 (materials applied: NM-101, P25)
Digestion of nano-TiO_2 in sediment and soil samples
For determination of titanium concentrations in sediment or soil the material was dried at 105 °C and then homogenized using a Retsch RM 0 milling device. Afterwards an exactly weighed amount of the anhydrous solid was digested in the UltraClave digestion system with conc. sulfuric acid. The required amount of sulfuric acid depends on the amount of TiO_2 in the sample. The digestion conditions of the microwave system were: heating up to 230°C in 30 min, then holding 230°C for 30 min. Thereafter the mixture was carefully brought to an exact volume with ultra-pure water, centrifuged for 30 min, 3200 rpm at 20°C and the supernatant was taken.

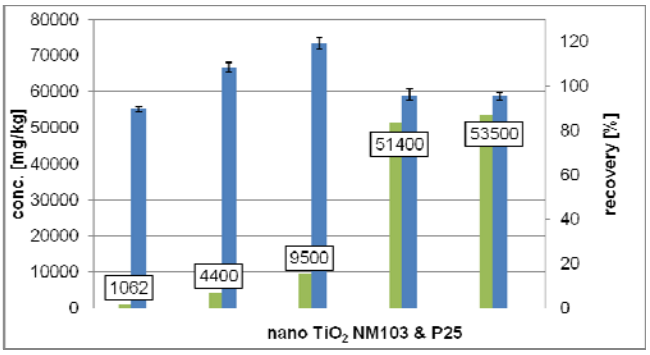

Figure 4: Sediment/soil spiked with known amounts of nano-TiO_2 NM103 and P25 (left scale; green bars; nominal concentration values are printed in the diagram) and recoveries after digestion (right scale)

Digestion of nano-TiO_2 in earthworms (*Lumbricus terrestris*)
Homogenization and lyophilization procedure is described in chapter nano-Ag / digestion of earthworms. However, the microwave digestion program differs: initial pressure 40 bar, heating up to 250°C in 60 min, then holding 250 °C for 30 min. After digestion 0.5 mL of hydrofluoric acid (40%) was added and the vessels were sonicated for 60 min. Strict safety precautions have to be considered for handling of hydrofluoric acid. Prior to measurement the samples were filled up to an exact volume with 4% boric acid to complex the fluoride ions.

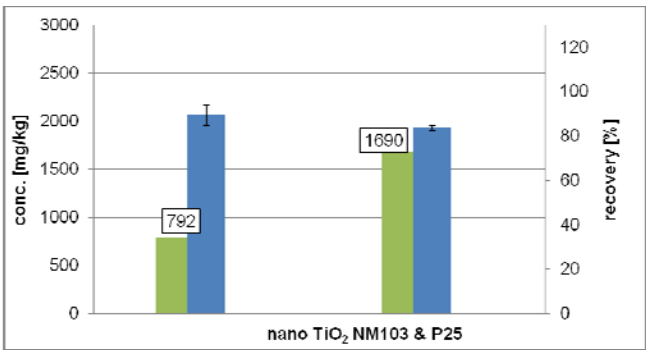

Figure 5: Earthworm samples (*Lumbricus terrestris*) spiked with known amounts of nano-TiO_2 P25 (left scale; green bars; nominal concentration values are printed in the diagram) and recoveries after digestion (right scale).

Digestion of nano-TiO_2 in aqueous samples

For fast dissolution of test solutions containing nano-TiO_2 samples were vigorously shaken for 5 min. Thereafter 1 mL of a mixture of 30% hydrochloric acid, 69% nitric acid and 40% hydrofluoric acid in a ratio of 3:1:1 was added to 4 mL of the sample. The mixture was sonicated for 30 min at 20°C. Afterwards boric acid was added for fluoride complexation.

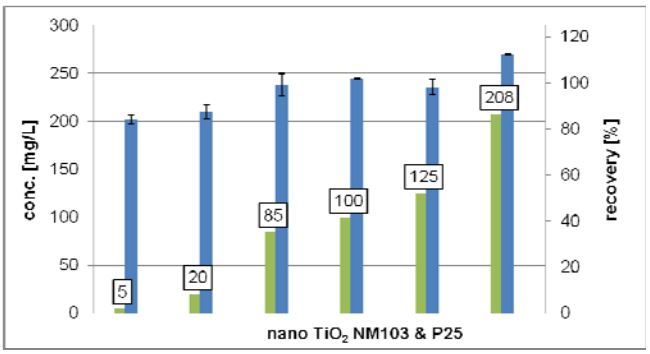

Figure 6: Aqueous medium spiked with known amounts of nano-TiO_2 NM103 or P25 (left scale; green bars; nominal concentration values are printed in the diagram) and recoveries after digestion (right scale).

Analytical quantification of Ti

Nano-TiO_2 reference materials with certified amounts of Ti were not commercially available at the time of development and investigation. Therefore, to validate the methods, chosen matrices were spiked with known amounts of nano-TiO_2, digested, quantified and their recoveries determined (Fig. 4-6). All final solutions were measured with matrix adjusted calibrations by ICP-OES (IRIS Intrepid II) or ICP-MS (Agilent 7500ce or 7700) for their amount of titanium. Commercially available Ti ICP standards were applied for calibrations of the instruments.

nano-Au (material applied: RM 8011)

Digestion of nano-Au in sediment samples

Digestion of nano-Au in dried sediment was performed following ISO11466 [3] using aqua regia. After cooling the mixture was carefully brought to an exact volume. The extract was filtered (supor membrane) and the filtrate used for quantification of Au.

Dissolution auf nano-Au in aqueous samples

A complete dissolution of nano-Au was achieved by mixing aqua regia and the aqueous sample in a volumetric ratio of 1:1. The mixture was prepared at least 8 hours prior to analysis.

Analytical quantification of Au

The reference material RM8011 (citrate-stabilized gold nanoparticles in aqueous suspension) with an information value for nano-Au was dissolved and quantified (appropriately diluted) to determine the recoveries (Fig. 7). All final solutions were measured with matrix adjusted calibrations by ICP-OES (IRIS Intrepid II, Thermo Scientific).

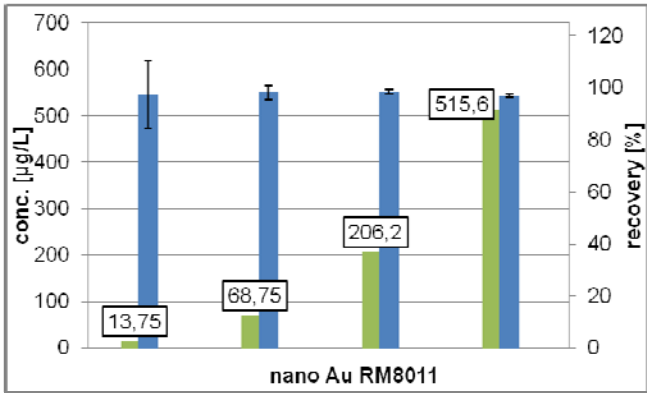

Figure 7: Recoveries (right scale) of appropriately diluted nano-Au reference material 8011 (left scale; green bars; nominal concentration values are printed in the diagram) after dissolution.

Conclusions

For the accompanying analysis of nanoparticles in ecotoxicological and fate tests the quantification of added nanoparticles is often a major task. We developed and implemented methods for complete dissolution or digestion of Ag-, TiO2- and Au- nanoparticles in different matrices and tested their performance. To assure the applicability of these methods different matrices were spiked with metal nanoparticles in different concentrations. After digestion or dissolution analytical recoveries were determined which were in the range of 100 + 25 %.

References

[1] http://www.understandingnano.com

[2] Handy, R. D., Cornelis, G, Fernandes D, Tsyusko, O., Decho A, Sabo-Attwood T., Metcalfe C., Steevens J. A., Klaine, S. J., Koelmans, A. A., Horne, N. 2012. Ecotoxicity test methods for engineered nanomaterials: practical experiences and recommendations from the bench. Environ Sci Technol 31, 15-31.

[3] Soil quality - Extraction of trace elements soluble in aqua regia. ISO 11466:1995, Beuth.

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