ESRF	**Experiment title:** Speciation of CeO_2-based nanocomposite within aquatic mesocosms	**Experiment number**: 30-02 1052
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Shifts: 12	**Local contact(s)**: Isabelle Kieffer	*Received at ESRF:*

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Report:

Introduction

In the past decade, there has been a growing concern about the use of nanoparticles/nanomaterials which are now present in 1000+ consumer products. Particularly, the high oxygen storage capacity of ceria NPs (CeO_2) makes them attractive for a large range of application like oxidation catalysts, gas sensor, polishing materials, but also as UV absorber as paint additive. This property is related to the easy Ce^{III}/Ce^{IV} redox cycle associated to the presence of oxygen vacancies in CeO_2. However, toxicity of CeO_2 NPs has been demonstrated for living organisms and the reactivity is induced by the presence of Ce^{III}. So, attractive technological impact due to the presence of Ce^{III} within structure of CeO_2 has to be considered together with potential environmental consequences after release in the environment.

To date, most of the research done on the environmental risk of NPs only concerns the hazard characterization, and rarely the exposure assessment. Moreover, the nano(eco)toxicological studies are mostly based on one kind of NPs exposed to one biological species in standardized and artificial media (*e.g.* micro-organisms, vertebrates, or invertebrates). Within the ANR-P2N MESONNET project, a mesocosms network was designed to assess the environmental risk of nanotechnology within environmentally relevant conditions. With these mesocosms, we study the distribution, transformation, and bioaccumulation of NPs within complex systems and working with environmentally relevant concentrations ($\leq$ 1 mg/L).

Experimental details

By combining different analytical approaches (e.g. µXRF, XRD, ICP-MS, DLS) we have identify the preferential zones of NPs accumulation in sediments and organisms from the mesocosms. This XAS experiment at the Ce L_3-edge consisted in determining Ce atomic environments in the different compartments following a single injection of 1 mg/L or 10 chronic injections of 0.1 mg/L of CeO_2-NPs. We evaluated the exposure and impact of two CeO_2-NPs: a surface modified CeO_2-NPs with citrate moieties used as paint additives (Nanobyk®), and an uncoated CeO_2-NPs (Rhodia®).

Ce L_3-edge XANES measurements on samples and reference compounds were performed at 5 to 10 K (helium cryostat), to avoid any possible dehydration or oxidation of samples. They were carried out on beamline BM30B/FAME with a Si(220) crystal monochromator operating at 6 GeV and 90 mA. The XAS spectra were all recorded in fluorescence mode, using a thirty-element solid-state germanium detector (Canberra, CT, USA). Surficial sediments, digestive glands (DG) of freshwater molluscs (*Planorbus* *corneus*), egg-layings, and juveniles were freeze-dried before XAS analysis. Individuals from the 3 replicates were mixed so that the amount of sample was enough for the XANES analysis, and to take into account the diversity between the 3 replicated mesocosms.

Results

Ce L_3-edge XANES analyses of *P. corneus* were performed to determine the speciation of Ce after ingestion or adsorption on the shell of the organisms. Both bare CeO_2-NPs and coated CeO_2-NPs are initially composed by Ce^{IV}. This is confirmed by their XANES spectra characterized by 2 absorption peaks at 5730.0 eV and 5738.0 eV while Ce^{III} reference compounds are characterized by one absorption edge at 5726.2 eV (Fig. 1). Ce in the DG of adults *P. corneus* possess a Ce^{III} signature for both bare and coated CeO_2-NPs (Fig. 1A). LCF of XANES spectra with initial NPs and Ce^{III}-cysteine reference compounds indicated that after 4 weeks, 78±1 % (Table 1) of Ce is reduced into Ce^{III} in the DG. The reduction of Ce^{IV} into Ce^{III} occurred less than one week after exposure. Surficial sediment was also analyzed by XANES and Ce^{IV} was mainly present in sediments during the 4 weeks of experiments with minor contribution of Ce^{III} (Fig 2).

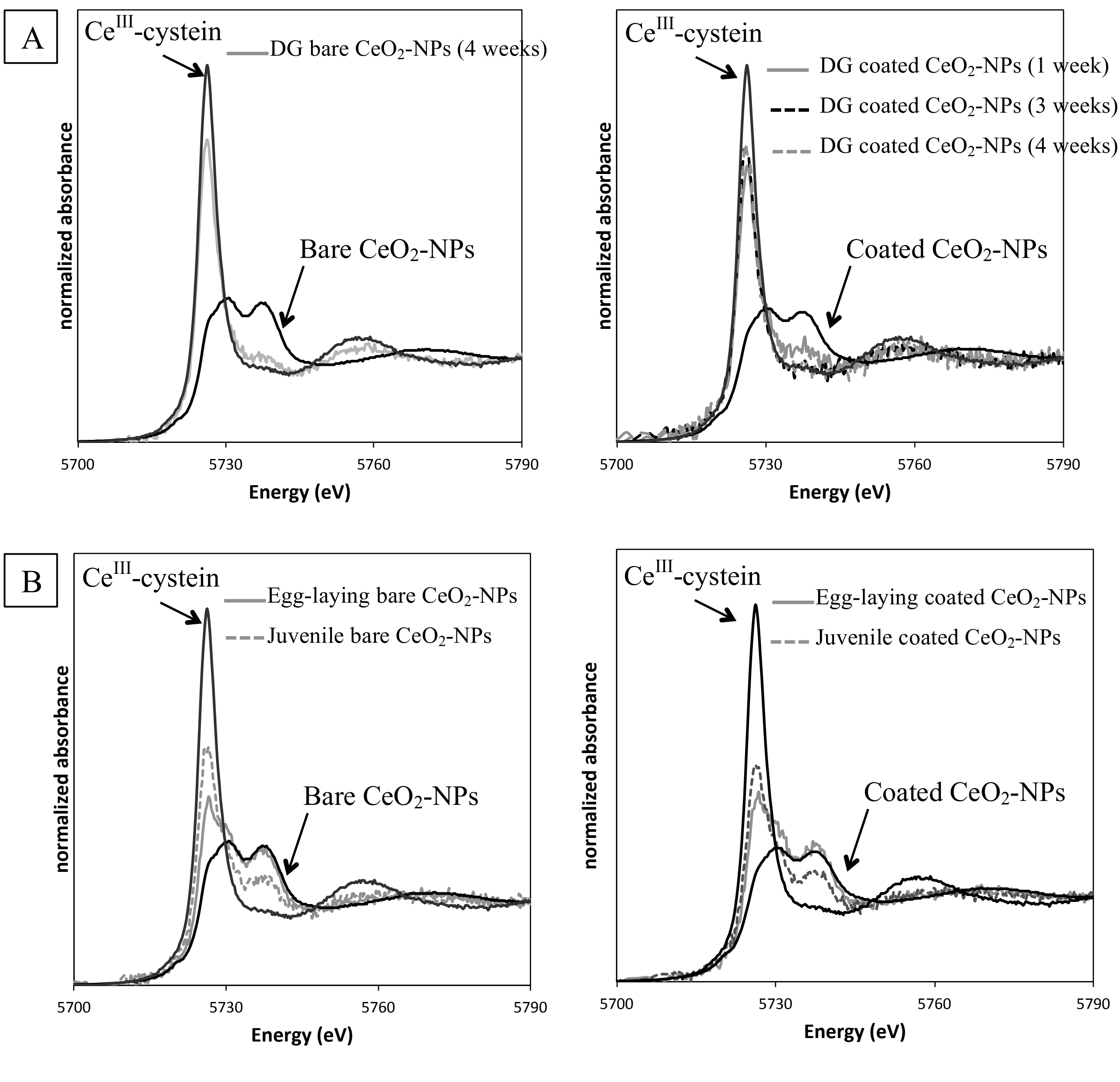

Figure 1. Ce L_3-egde XANES spectra in *P. corneus*. A/ DG from adult P. corneus exposed to bare and coated CeO_2-NPs at different period of time. B/ Egg-laying (sampled after 1 week) and juvenile (sampled after 4 weeks). Unknown samples are compared to bare and coated CeO_2-NPs, and Ce^{III} reference spectra.

For juvenile *P. corneus* and egg-layings, the reduction into Ce^{III} was less important (Fig. 1B). For juvenile, the LCF indicated that 47±5% of the Ce^{IV} is reduced into Ce^{III} after 4 weeks (Table 1). The egg-layings were sampled after 1 week and only 22±1% of the Ce was reduced into Ce^{III}. No significant difference in Ce speciation was observed on the DG of adults, juveniles, and egg-layings as a function of the type of NPs.

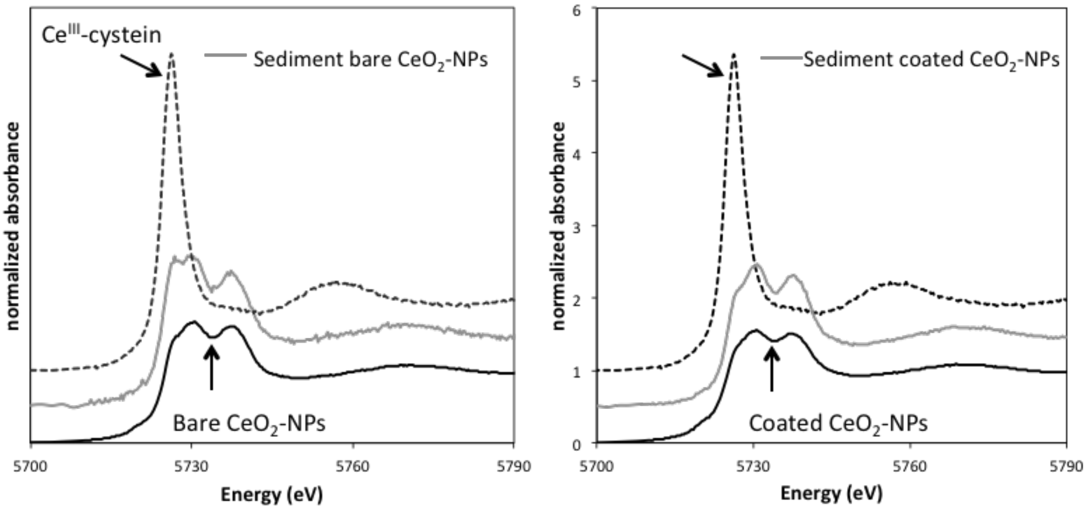

Figure 2. Ce L_3-egde XANES spectra in surficial sediments after 4 weeks compared to bare and coated CeO_2-NPs, and Ce^{III} reference spectra.

XANES analyses were performed on all samples envisaged. XAS results were put in perspective with oxidative stress data to evaluate the fate and impacts of CeO_2-NPs within aquatic ecosystems. Two publications based on the data acquired during this XAS session are currently in progress.

Table 1. Speciation of Ce bioassimilated by *P. corneus* (DG of adults, laying and juveniles) determined from Ce L_3-edge XANES experimental sepctra and their linear combinaison fitting (LCF) with XANES spectra of reference materials (Ce^{III}-Cysteine complex and CeO_2-NPs).

	Coated CeO_2-NPs			**Bare CeO_2-NPs**		
Digestive glands	Ce^{IV}	Ce^{III}	R-factor	Ce^{IV}	Ce^{III}	R-factor
1 week	35%	65%	0.004	NA	NA	
3 weeks	20%	80%	0.006	NA	NA	
4 weeks	21%	79%	0.007	23%	77%	0.002
Laying after 1 week	77%	23%	0.006	79%	21%	0.002
Youngs after 4 weeks	58%	42%	0.002	49%	51%	0.002

Communications:

23th Goldschmidt Conference 2013, 25-30 August 2013, Florence, Italy. **M. Tella**, et al., Environmental fate and impacts of ceria nanomaterials: distribution, transformation and bioaccumulation within aquatic mesocosms (oral presentation).

Nanosafe 2012, 13-15 November 2012, Grenoble, France. **M. Tella**, M. Auffan, et al., Assessment of environmental exposure to nanomaterials through mesocosms experiments (oral presentation).