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Report:

Introduction

Surface layers (S-layer) are highly ordered protein layers on the surface of many bacteria and possess a high potential to bind metals. In this work interactions between Pt- and Pd-nanoclusters and the S-layer protein of the uranium waste pile isolate *Bacillus sphaericus* JG-A12 and the reference strain *B. sphaericus* NCTC 9602 were studied to identify the chemical groups of the protein which were responsible for the binding of the metal nanoclusters. Additionally the nanocluster formation during reduction of the metals should be confirmed. The latter was demonstrated with HR-TEM examinations.

Experimental procedure for EXAFS sample preparation

The S-layer samples (with a protein content of 3.9-9.3mg/ml in 50mM $\text{KH}_2\text{PO}_4/\text{Na}_2\text{HPO}_4$, 3mM NaN_3 , 1mM MgCl_2 pH 7.5) were prepared by adding a 24h

old 30mM K_2PtCl_4 and K_2PdCl_4 solution and incubation for 24h in darkness. Using dialysis and centrifugation the protein with bound metal nanoclusters was separated from surplus K_2PtCl_4 - or K_2PdCl_4 -solution, resuspended in water, dried in a vacuum incubator and grounded. Assuming that 50% of K_2PtCl_4 or K_2PdCl_4 was reduced to Pt or Pd clusters, 2 to 8 mg platinum or palladium should be bound to the S-layer. Reference samples were prepared with 50mM KH_2PO_4/Na_2HPO_4 , 3mM NaN_3 , 1mM $MgCl_2$ pH 7.5 buffer instead of a protein buffer solution

EXAFS measurement

The EXAFS measurements were carried out at the Rossendorf Beamline (ROBL). Platinum L_{III} -edge and Palladium K-edge X-ray absorption spectra of the dry samples were collected in transmission and fluorescence mode.

Results

In all Pd samples the Fourier transformation (FT) of the first dominating peak shows a amplitude function, which is typical for oxygen or nitrogen. Using Pd-O phase and amplitude functions, the FT gives a distance of $1.99\text{-}2.02\pm 0.02$ Å for the Pd-S-layer samples. This bond length is in good agreement with the distance between palladium and oxygen in PdO (2.018 Å). The reference sample of pure Pd-clusters shows an additional peak at a distance of 3.06 ± 0.02 Å originated by Pd-Pd interaction. The equivalent but less distinctive peak the Pd-S-layer samples give distances of $3.03\text{-}3.04\pm 0.02$ Å. This distance differs significantly from the distance expected for the Pd metal ($R=2.758$ Å), but is in good agreement with the Pd-Pd distance in PdO ($R=3.03$ Å). For this reason the dominating species in all samples seems to be oxidized. The latter makes it difficult to get information about the chemical groups, which are responsible for binding of the metal clusters. For the Pt samples similar results are obtained with the exception, that there is no clear Pt-Pt interaction.