Supplementary data

Sulfur K-edge XANES study of dihydrolipoic acid capped gold nanoparticles : dihydrolipoic acid is bound by both sulfur ends.

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Synthesis of dihydrolipoic acid (DHLA) capped gold nanoparticles (Au@DHLA). For a typical preparation of 5 nm gold particles, 4.10^{-5} mol of $HAuCl_4.3\ H_2O$, dissolved in 10 mL methanol were placed in a 100 mL round bottom flask. 8.10^{-5} mol of DHLA in 3.2 mL of methanol and 0.16 mL of acetic acid were added to the gold salt solution under stirring. The mixture turned from yellow to orange. After 5 minutes, 40.10^{-5} mol of $NaBH_4$ dissolved in 1.1 mL water were added to the orange mixture under vigorous stirring and at room temperature. At the beginning of the $NaBH_4$ addition, the solution became first dark brown then a flocculent black precipitate appeared.

The vigorous stirring was maintained for 1h before adding 5 mL of 1 M aqueous hydrochloric acid solution. After the partial removal of the solvent under reduced pressure and at maximum 40°C, the precipitate was filtered on a polymer membrane, washed with 0.01 M HCl, water and ether. The resulting black powder was dried and stocked in the solid state or dispersed in 10 mL of 0.01 M NaOH aqueous solution.

Size Characterization and ζ-potential measurements. Direct determination of the size distribution (in the range 1-100 nm) and of the ζ-potential of the Au@DHLA nanoparticles were performed via a Zetasizer 3000 HS (laser He-Ne (633 nm)) from Malvern Instrument. Prior to the experiment, the sol was diluted to obtain a concentration of around 0.08 $g.L^{-1}$ in an aqueous solution containing 0.01M NaCl and adjusted to the desired pH.

The hydrodynamic diameter (Dh) of Au@DHLA is mainly comprised between 5.1 and 8.0 nm (Figure 1S).

High Resolution Transmission Electron Microscopy. HRTEM was used to obtain detailed information about the samples and was carried out using a JEOL 2010 microscope operating at 200 kV. The samples for HRTEM were prepared by depositing a drop of a diluted Au@DHLA colloidal solution on a carbon grid (Figure 2S). The diameter of most nanoparticles is around 5 nanometer.

X-ray Photoelectron Spectroscopy (XPS). The surface chemical composition of Au@DHLA was determined by XPS (Table 1S). XPS analysis were carried out at the "Institut de Recherche sur la Catalyse" (IRC) with a VG Scientific ESCA LAB 200 R using monochromatic Al Kα X-ray sources (hν=1486.6 eV). The binding energies scales for the monolayers on gold were referenced by setting the $Au_{4f7/2}$ binding energy to 84.0 eV. Spectra were recorded at a takeoff angle of 35° (angle between the plane of the sample surface and the entrance lens of the analyzer) and with a pass energy of 50 eV. The theoretical analyzer resolution expected with that setting is 1.5 eV.

As expected, the experimental atomic composition of the adsorbed organic layer is close to the one calculated for DHLA. The main explanation of the too high value of the experimental

C/S ratio lies in the contamination of sample. Indeed the contamination by environmental hydrocarbons is almost unavoidable.

X-ray Absorption Near Edge Spectrocopy (XANES) set-up. This work was carried out at the X-ray Microscopy Beamline (ID21) of the European Synchrotron Radiation Facility (ESRF) in Grenoble (France), which gives access to the 2-7 keV energy range. The Scanning X-ray Microscope (SXM) of ID21 offers an attractive tool for micro-spectroscopy at the sulfur K-edge. For this experiment, the SXM was operated at energies around 2472 eV, which corresponds to the sulfur K-edge. The harmonics rejection was ensured by a set of two parallel silicon mirrors deflecting in the horizontal plane with an incident angle of 8 mrad. The energy scan was performed by a fixed-exit double crystal Si(111) monochromator providing a spectral resolution of 0.5 eV in this configuration. This high energy resolution is essential to resolve the required sulfur K-edge XANES features. In focused configuration, Fresnel zone plates are used as focusing optic in the SXM, demagnifying the X-ray source to generate a submicron probe, typically 0.3 x 0.3 μm^2 with a photon flux of 4 x 10^8 photons.s^{-1} at the sulfur K-edge. However, for this experiment which did not require spatial resolution, the SXM was used in unfocused mode, i.e. simply using a 500 μm pinhole defining the beam size. The sample stage was tilted by 30° with respect to the beam direction and the fluorescence photons emitted by the sample were collected either by an energy-dispersive high purity germanium detector (Princeton Gamma-Tech) for the samples exhibiting low S concentration, or by a Si photodiode for the more concentrated ones. The fluorescence detectors were placed perpendicular to the beam direction to minimize scattering effects. The monitoring of the incoming beam intensity on the sample, which is essential to correct the acquired XANES spectra and images for beam intensity fluctuations, was ensured using a drilled photo-diode collecting the fluorescence from a 0.75 μm thick Al foil inserted in the

beam path. In order to avoid the strong absorption of the X-ray incoming beam and the sulfur emission lines by air, the SXM chamber was used under primary vacuum (10^{-4} mbars).

ENERGY CALIBRATION

Spectra of sulfur-containing reference compounds were recorded first. The latter are used both for energy calibration of the monochromator and as fingerprints for assigning the resonances observed in the real samples. The standards used were cystine, cysteine, glutathione, methionine and chondroitin sulfate. In the case of L-cystine (Sigma), the energy positions of the two main resonances were found at 2472.4 eV and 2474 eV. The energy was scanned between 2450 eV and 2520 eV in 0.175 eV energy steps, with a dwell time varying between 1s and 10s for the less concentrated samples. To follow up possible photo-reduction effects due to the X-ray irradiation, large numbers of fast energy scans were performed and summed for each spectrum. No photo-reduction effect was evidenced.

XANES data processing

Spectrum processing was performed in the following way. First, a linear background determined in the pre-edge region was subtracted from the spectrum to correct for the absorption introduced by higher shells and other absorbing atoms than the central S atoms. The edge jump of the spectra was then normalized to unity at 2510 eV, where the effects of the strong near edge resonances become small.

An arctangent step function was used to model the transition of photoelectrons to continuum, and Voigt functions were used to fit the near edge features. Voigt line shape which is a mixture of Lorentzian and Gaussian functions was selected to account for the Lorentzian shape of the transitions convoluted by the Gaussian broadening introduced by the beamline optics (Rose *et al.*, *J. Am. Chem. Soc.*, 1998, **120**, 10743. ; Farges *et al.*, *J. Non-cryst. Solids*,

2004, in press). Least square fitting was performed with Peak Fit (Figure 3S).

XANES spectra distortion

The demonstration of the DHLA anchorage on gold thanks to both sulfur ends was performed by comparing the position of the main peaks. Such an interpretation is reliable only if the position of the peaks was not sensitive to the self-absorption which induces spectrum distortion. The self-absorption effects occur for thick or concentrated samples (i.e. with sulfur content higher than 0.3% in weight ; Waldo *et al.*, *Cosmochim. Acta*, 1991, **55**, 801. Xia *et al.*, *Soil Sci. Soc. Am. J.*, 1998, **62**,1240). Among the seven XANES spectra, described in the manuscript, three out of them (pure TA, pure DHLA and Au@DHLA in the solid state, Figures 1a-1c in the manuscript) can potentially be distorted because of the high sulfur content (~15% for TA and DHLA and 1.5% for Au@DHLA in solid state). The sulfur content of the Au@DHLA particles (~0.006%) dispersed in an alkaline aqueous solution is largely smaller than 0.3% in weight.

To evaluate the distortion effects on the peaks position, XANES spectra of thick and thin layers of TA were performed in fluorescence and compared with that of thin layer of TA obtained by transmission measurements which should be distortion free (I.J. Pickering *et al.*, *Biochemistry*, 2001, **40**, 8138). Thioctic acid spectrum (Figure 4Sa corresponding to Figure 1a in the manuscript) was measured in fluorescence on a thick sample. To minimize self absorption effects, we performed the measurement again on a finely ground sample, deposited as a thin layer on sulfur free adhesive tape. Care was also taken that the layer be as homogeneous as possible to minimize pinhole effects which might affect transmission measurement of spectra on a sample with inhomogeneous thickness (Prange *et al.*, *Microbiology*, 2002, **148**, 267., and Microbiology comments : George *et al.*, *Microbiology*, 2002, **148**, 2267. and Prange *et al.*, *Microbiology*, 2002, **148**, 2268.) . The spectrum was

measured simultaneously in transmission and fluorescence mode (Figure 4S). The self absorption effect is visible as a damping of the near edge features, but still peaks are found at the same energy positions.

The absence of peak shift owing to the self-absorption is in agreement with the data collected by Pickering *et al* (I.J. Pickering *et al.*, *Biochemistry*, 2001, **40**, 8138).

Table 1S. Atomic compositions obtained from the area of core level photoemission peaks corrected by sensitivity factors (meas) or from stoichiometric ratios (calc)

	C/S		O/S	
	meas	calc	meas	calc
Au@DHLA	4.525	4.0	1.075	1.0

Figure 1S. Au@DHLA particles size distribution

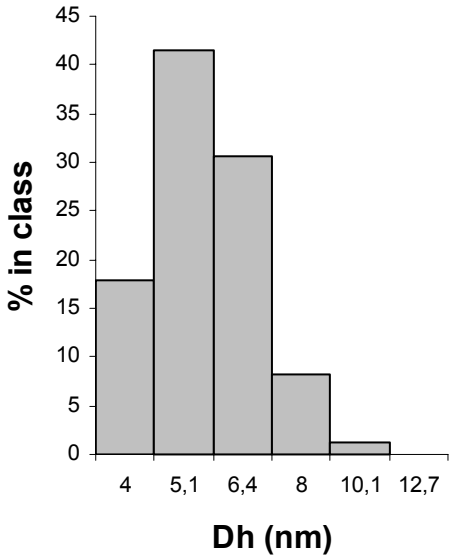

Figure 2S. HRTEM of Au@DHLA particles

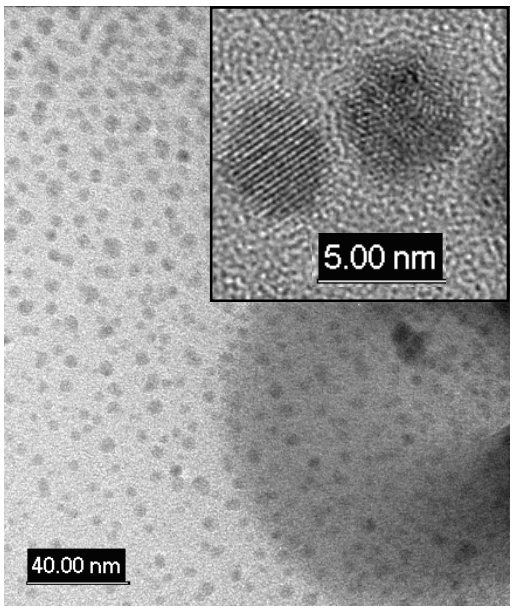

Figure 3S. Example of deconvolution of the XANES spectrum into Voigt and arctangent functions. Solid line represents the least-squares fitted spectrum, squares represent the experimental data. The deconvolution of the spectrum into pseudocomponents is shown at the bottom.

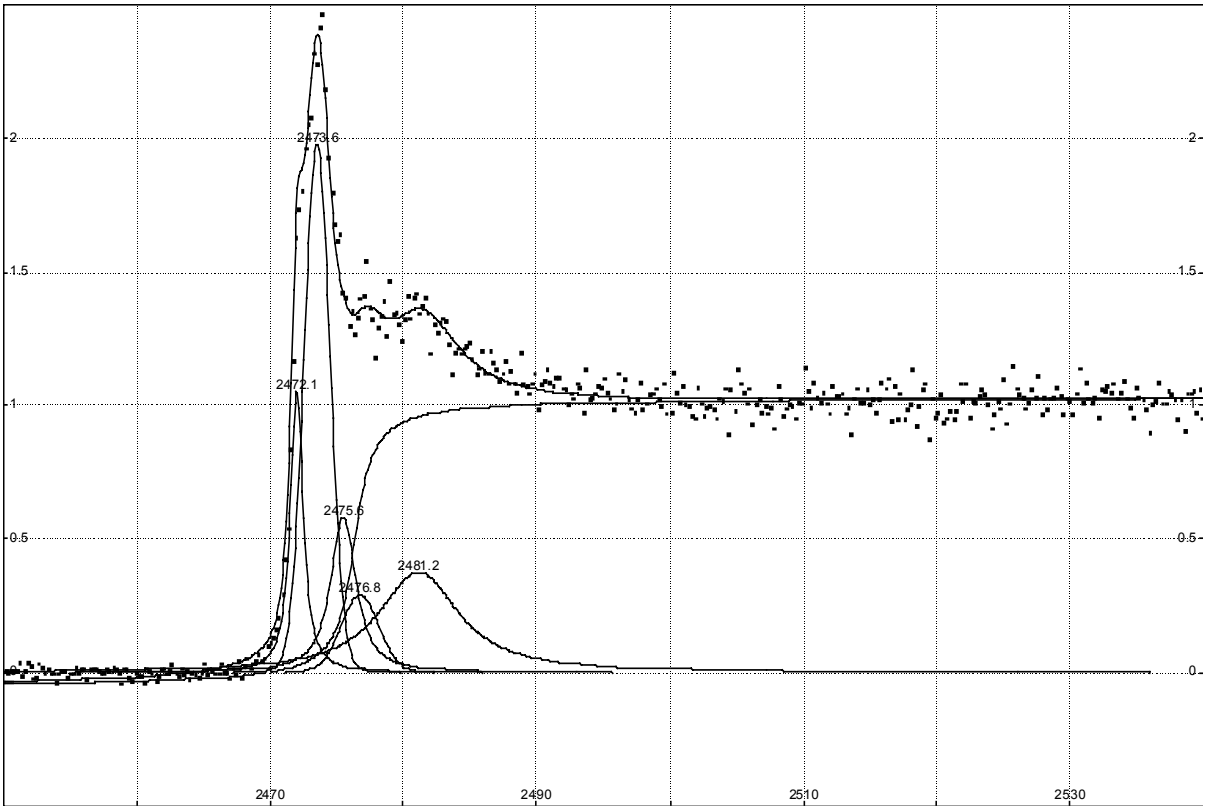

Figure 4S. fluorescence spectra of thick (a) and thin (b) thioctic acid samples and transmission spectrum of thin thioctic acid layer (c).