ESRF	**Experiment title:** Arsenic contamination in sediments, plant and animal species from a WWF reserve	**Experiment number**: EV-490
Beamline: BM 08	**Date of experiment**: from: 29/06/2022 to: 04/07/2022	**Date of report**: 07/07/2022
Shifts: 15	**Local contact(s)**: Francesco d'Acapito	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Chiara Petroselli*, PhD. Dipartimento di Chimica, Biologia e Biotecnologie, Università degli Studi di Perugia, Perugia (Italy)

Matteo Pallottini*, PhD. Dipartimento di Chimica, Biologia e Biotecnologie, Università degli Studi di Perugia, Perugia (Italy)

Federica Bruschi*. Dipartimento di Chimica, Biologia e Biotecnologie, Università degli Studi di Perugia, Perugia (Italy)

Eleonora Marchetti. Dipartimento di Chimica, Biologia e Biotecnologie, Università degli Studi di Perugia, Perugia (Italy)

Report:

Experiment EV-490 was designed to investigate arsenic (As) speciation in various environmental matrices (lacustrine sediments, plant and animal tissues) sampled at the WWF reserve "Oasi di Alviano" in Central Italy. The study was aimed at determining As speciation in various organs of selected plant and animal species to understand their coping mechanisms with this toxic metalloid and its localisation inside the living organism. Measuring sediments, plant and animal samples in the same experimental session also allows to follow the propagation of the element of interest alongside the food chain and in various environmental compartments (aquatic, terrestrial).

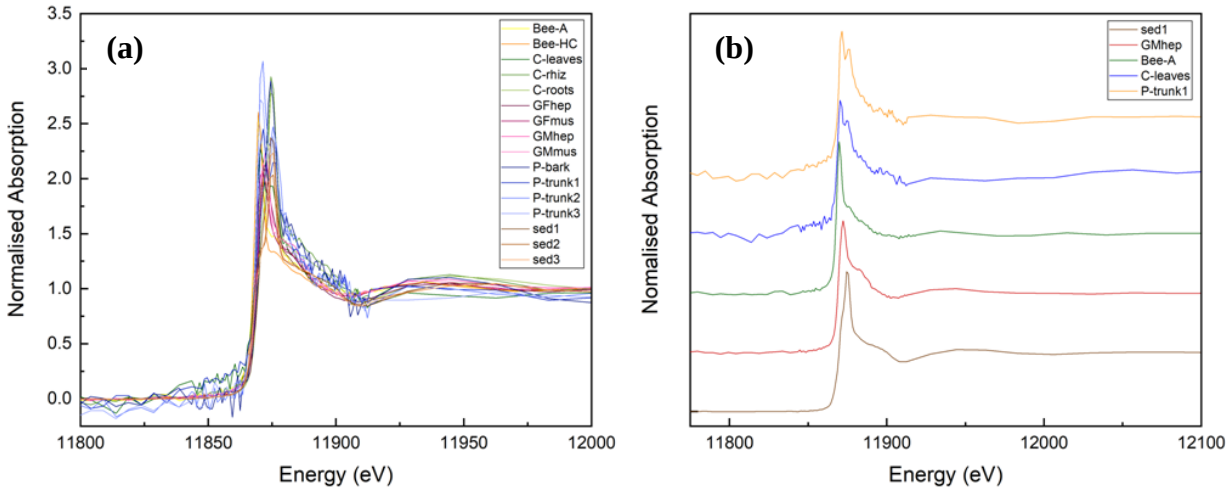
Background. The "Oasi di Alviano" WWF reserve is part of the Natura 2000 Network as Special Protection Area (IT5220024) and Special Area of Conservation (IT5220011), and it's one of the world's largest WWF reserves, hosting all of the wetland habitats and hundreds of plant and animal species. Despite the area is protected since 1990 and therefore it doesn't have any direct local pollution sources, arsenic and mercury contamination was detected in the samples collected in this area. The source of these toxic elements can be identified as the mercury mines on the Mount Amiata situated in Tuscany, upstream of the Oasi. This area is part of the global mercury belt and its mines have been exploited for many years. Although the mining site was dismissed 30 years ago, the contaminated fluvial sediments still act as a diffuse pollution source as they have been transported all the way through the Paglia basin to the Tiber basin and finally to the Alviano area.

Samples treatment. The samples analysed in this study were collected during multiple sampling campaigns at the "Oasi di Alviano" between 2020 and 2021. The 3 selected sediment samples correspond to the superficial, middle and lower parts of a 30 cm sediment core sampled in July 2021. Sediment samples were air dried, sieved at 2 mm, homogenised and milled. They were then further dried at 60°C for 24h and mixed with anhydrous cellulose to be pressed into a pellet for XAS analysis (d=13mm). Poplar plant samples included leaves, bark and a trunk core (outer, middle and inner part). Common reed plants were divided into roots, rhizomes, stem and leaves. All the plant samples were freeze-dried and grounded, and then pellets were made using 100% sample. Honey bees were divided into head-thorax and abdomen samples, while for red swamp

crayfish we analysed muscle tissue and hepatopancreas. All the animal samples were freeze-dried and grounded, then pellets were made using sample only (100%). The arsenic content in all of the samples was previously determined in the University of Perugia laboratories by means of quantitative analytical techniques (ICP-MS).

Experimental session. During the first day of beamline setup and preliminary measurements, we have faced the difficulty of detecting the As signal in the X-ray fluorescence spectrum for most of the samples. We have therefore dedicated one day to acquiring test XANES spectra in order to explore more in deep the possibility to analyse our samples (verifying we could acquire an edge jump). We demonstrated that it was possible to acquire a XANES spectrum for most of the samples (except for poplar leaves and reed stem), however, a full EXAFS spectrum was obtainable for sediments and reed roots only. Following the main scope of the experiment, we therefore decided to focus on the XANES part of the spectrum and to extend the acquisition time in order to reduce the noise. Since the storage ring refill would affect the quality of the spectra, we have synchronised the spectrum acquisition (53 minutes) with the refill times (every 1 h) and we increased the number of scans (4 to 7 scans per sample, depending on the signal strength). All the samples were recorded in low vacuum at room temperature, in fluorescence mode, however, different working conditions were required for adjusting the signal of different types of samples (e.g. Al filter in front of the detector for sediment samples rich in Fe). We have also recorded XANES spectra for six standard As compounds, in the same conditions as the samples but in transmission mode. The transmission spectrum of a reference sample (GaAs) was recorded at the same time as each sample scan in order to provide a reliable internal energy calibration. The new experimental setup with a Si(111) crystal monochromator and an SDD multi-element detector array (ARDESIA,1) ensured the possibility to analyse very diluted samples. Moreover, in order to explore the possibilities of the new beam size (100 µm) for future proposals, we have also dedicated one shift to mapping a whole honey bee and recording the XANES spectra in selected points of the body. We therefore demonstrated the feasibility of these type of measurements, opening the way to new applications in the investigation of metals speciation and distribution inside an organism.

Results. During the experimental session, we were able to record the XANES spectra at the As k-edge (11867 eV) for 3 sediment samples, 3 common reed samples (roots, rhizome, leaves), 4 poplar samples (bark, trunk1-3), 2 honey bee samples (head/thorax, abdomen) and 4 red swamp crayfish samples (muscle and hepatopancreas, male and female). All the spectra are shown in Figure 1a. Despite the higher number of scans performed for more diluted samples, the quality reached is not the same as that for more concentrated samples (Figure 1a,b). No differences in As speciation were recorded for the 3 poplar trunk samples which could then be averaged together to further reduce the noise. On the other hand, differences in As speciation were observed between plant and animal samples, as preliminarly shown in Figure 1b. The obtained data, thus, show promise for the interpretation of the As speciation evolution along the food chain.



References:

Bellotti, G. et al. 2018 *"The ARDESIA Detection Module: a 4-Channel Array of SDDs for Mcps X-Ray Spectroscopy in Synchrotron Radiation Applications"* IEEE Transactions on Nuclear Science, Vol. 65, NO. 7, DOI: 10.1109/TNS.2018.2838673