

Study of arsenic uptake in cucumber by XANES at HASYLAB Beamline L

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Different speciation of elements strongly influences the metabolic processes in biological systems depending on their chemical states. The arsenic is a toxic element for biological systems, that element occurs frequently in the nature. The toxicity level of the arsenic compounds is determined by the oxidation state of the arsenic. In this project we have studied the uptake effect of cucumber plants as a model biological system in order to examine the accumulation of toxic elements: the distribution of arsenic in cross section of cucumber hypocotyls was determined at HASYLAB DORIS-III Beamline L in lyophilised samples with confocal X-Ray fluorescent spectrometry [1]. Furthermore, to obtain information about the chemical form of the arsenic content of the samples X-Ray absorption near edge structure (XANES) measurements under “in-vivo” conditions were carried out in fluorescent detection mode at Beamline L. The oxidation state of the arsenic accumulated in the hypocotyl should change during the sample preparation procedure especially at the case of lyophilisation. To avoid this influence on the chemical status of the arsenic living cucumber plants were measured. Due to the damage in the biological structure of the living sample caused by the high X-ray flux of the synchrotron beam cryogenic cooling was applied with LN₂ steam. The cryogenic setup does not allow applying polycapillary due to the high risk of damage of the optical element. Silicon 111 crystal was used to monochromatize the synchrotron white beam. In order to neglect the damage of the sample caused by the intense excitation flux the size of the beam was set by slits with size of ≈ 1.0 mm. Two arsenic standard solutions were measured to obtain the pure As(III) and As(V) absorption spectra (Figure 2.). The energy resolution was 0.2 eV with 1 s measuring time without liquid nitrogen cooling.

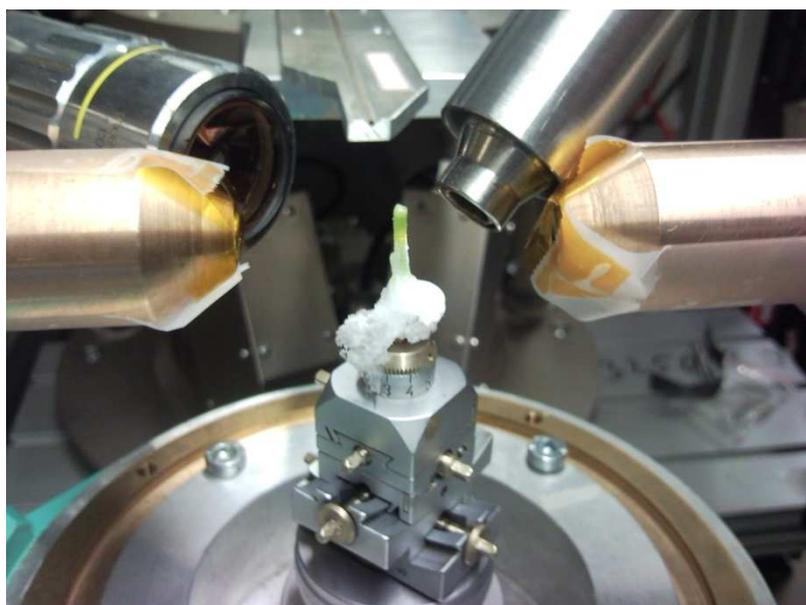


Figure 1. XANES measurement set up with cryogenic cooling by LN₂ stream.

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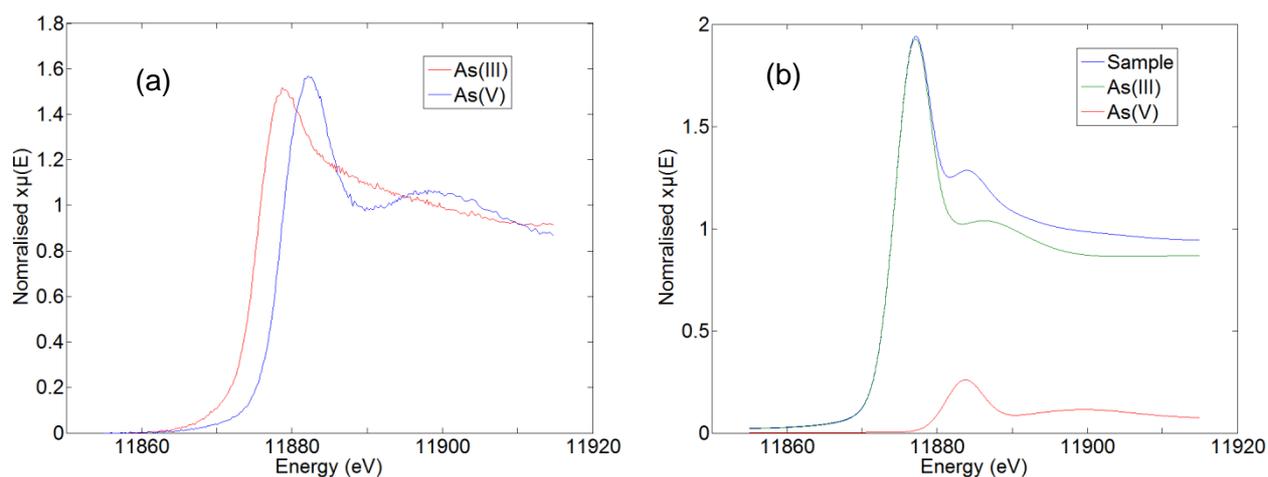


Figure 2. (a) XANES spectra of the As(III) and As(V) standard samples and (b) the result of the fitting on spectra of hypocotyl sample.

The cucumber plants were grown up in standard nutrient solutions and they were poisoned with solution of different arsenic concentrations (40, 60, 80 and 100 μMol). Living cucumber hypocotyl samples were measured by such a way, that the cooling jet of liquid nitrogen steam was oriented to just below the measured sample area, i.e. the irradiated volume of the sample was not cooled directly. With this cooling mode the level of the ice formation was strongly limited on the irradiated area because the heat conductivity of the sample body was enough for cooling the irradiated part of the sample. Repeated measurement of the fluorescent signal emitted by the arsenic of the same sample resulted constant values, therefore the chemical states of the arsenic did not change during the irradiation period and we could not recognised optically any beam-damage as well. The measuring time was set between 5-10 s depending on the actual intensity of the arsenic fluorescence signal.

First the spectra of standard solutions were fitted using an own developed software in MATLAB environment [2]. At the case of the real samples the ratio of the two type of arsenic oxidation states (As(III) and As(V)) were determined by fitting their XANES spectra with the linear combination of the fitted spectra of the two standard solutions. Based on the result of these fits, the relative concentrations of the two arsenic chemical forms were determined in the cucumber hypocotyl samples.

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