Confocal X-ray fluorescence micro-imaging on cucumber hypocotyls

A. Gerényi[1], V. Czech[2], J. Garrevoet[3], B. De Samber[3], L. Vincze[3] and I. Szalóki[1]

[1]Institute of Nuclear Techniques, Budapest University of Technology and Economics, 1111 Budapest, Hungary
[2]Department of Plant Physiology and Molecular Plant Biology, Eötvös Loránd University, 1117 Budapest, Hungary
[2]Department of Analytical Chemistry, Ghent University, B-9000 Ghent, Belgium

The toxicity in plants caused by environmental pollution by heavy metals such as lead, nickel etc. and other toxic elements such as As has been studied by confocal X-ray microfluorescence. In many cases plant tissues accumulate these toxic elements and transport them in the xylem and phloem sap from roots to the leaves. The toxic elements and compounds strongly influence the metabolic processes; however some plants develop an efficient mechanism against this phytotoxicity [1]. Some plants may therefore constitute aconsiderable risk factor for human health when entering the food chain. Such a typical toxicity problem is caused by arsenic which can be taken up by most of the plants and this effect can be used as indicator on the bioavailability in the soil.

To obtain information on the ion-transport mechanisms in cucumber plants, micro-XRF measurements in confocal geometry were performed on cucumber hypocotyls. A high flux polycapillary was used as a focusing device for the primary beam with a high transmission efficiency and long focal distance (≈ 8 mm). The diameter of the focal spot size was measured to be 15-20 μm as a function of the primary energy. The confocal polycapillary optic mounted in front of the SDD detector with a shorter focal distance (≈ 2.5 mm) provided an acceptance of approximately 15 μm at the As-Kα energy. The focal spot at the sample position was determined to be 13x17 μm (FWHM) at 14.0 keV. The applied experimental arrangement is shown in Fig. 1. The samples were lyophilized in order to reduce beam-damage effects typically caused by the intense focused beam in the living sample cells. The photo on Figure 2.b. demonstrates the beam damage effect on a living sample after 300 s irradiation. The arsenic concentration in the nutrient solution used for the cucumber culturing was set to 80 μMol/dm^3. In order to achieve an optimum value for the arsenic XRF signal the excitation energy was set to 12.5 keV using the multilayer monochromator, which is just above the arsenic K-edge energy providing maximum excitation efficiency.

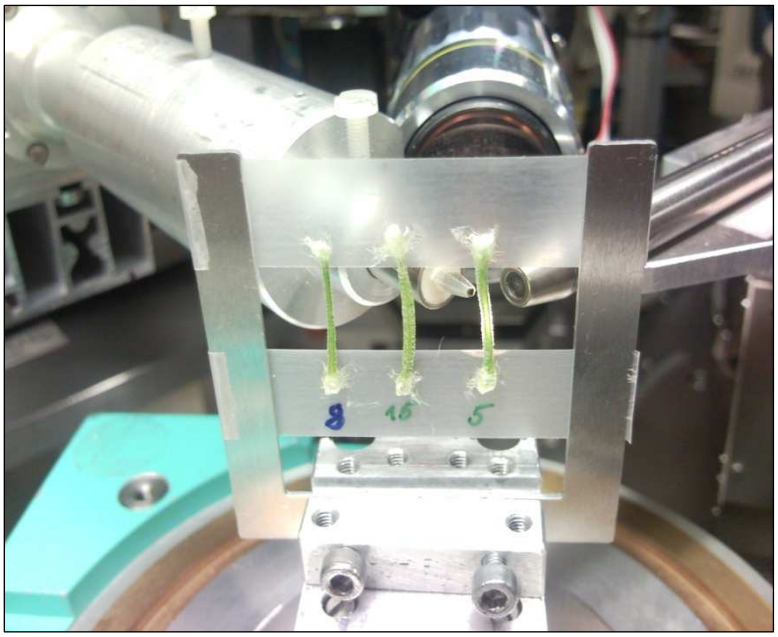

Figure 1. Measuring set-up for μ-XRF confocal imaging performed on cucumber

Confocal 2D micro-XRF maps were taken from given horizontal cross-sections across thecucumber hypocotyl's (see As-Kα distribution in Fig.1a). In order to achieve the highest lateral resolution under the given experimental conditions the sweep scans were performed

using a 10 μm step size (i.e. voxel size). The sampling time per voxel was set in the range of 5-10 seconds, which condition allowed reaching a few hundred cps/point count-rate for the arsenic Kα signal.

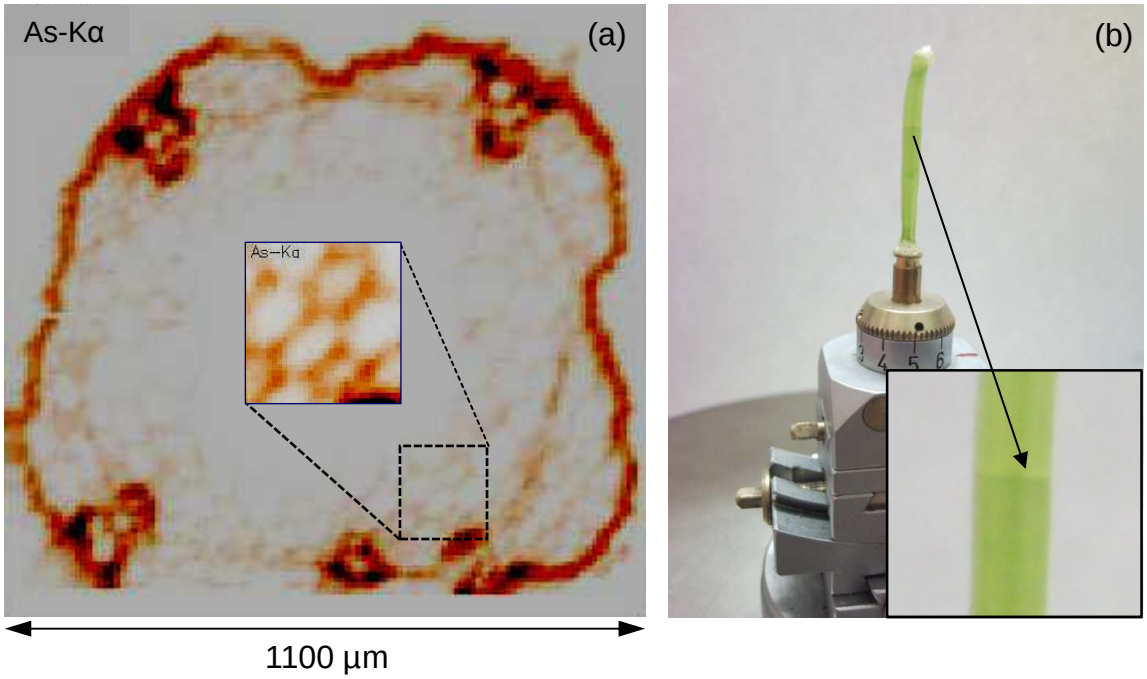

Figure 2. (a) As-Kα intensity distribution in a hypocotyl cross-section and (b) beam-damage effect in a living sample caused by the excitation

The results of the confocal XRF imaging shows that the As was transported into the individual cells of the hypocotyls. The sizes of these special (giant) cells in the hypocotyls are approximately 50-100 μm. The xylem channels located in the corners of the hypocotyls shown in Figure 2(a) are also rich in arsenic which indicates the conclusion: the entire cross-section of the studied cucumber plant exhibits high concentration levels of arsenic. In spite of the detected high concentration levels, our optical observation on living cucumbers confirmed that the plants tolerated well the arsenic shock caused by the relatively high As concentration in the nutrient solution. Finally, it can be concluded that the applied confocal micro-XRF setup was successful in the visualisation of trace-level As adsorption in plant cells by making use of (i) optimum excitation efficiency for the As-Kα line with the appropriate selection of the excitation energy and (ii) over-sampling the scanning step size.

References:

[1] V. G. Mihucz, G. Silversmit, I. Szalóki, B. Samber, T. Schoonjans, E. Tatár, L. Vincze, I. Virág, J. Yao, Gy. Záray, Removal of some elements from washed and cooked rice studied by inductively coupled plasma mass spectrometry and synchrotron based confocal micro-X-ray fluorescence, *Food Chemistry*, Vol. 121, pp. 290-297, 2010.

Acknowledgements

This work was supported by HASYLAB and it is partly connected to the scientific program of the „Development of quality-oriented and harmonized R+D+I strategy and functional model at BME" project. This project was partly supported by the New Hungarian Development Plan (Project ID: TÁMOP-4.2.1/B-09/KMR-2010-0002).