

Experimental report

XAS study of iron compounds in *Imperata Cylindrica* plant from Tinto River

Biominerals as iron is stored in plant cells in cavity ferritin proteins. While structural, chemical and magnetic properties ferritin in animals is well established, characterization of iron storing in plant cells systems remain on study. Here, we report a detailed work of composition, location of naturally formed iron nanocompounds cores in plant cells tissues grown in iron concentrated environments by means X-ray absorption (XAS) at the Fe K-edge, in both XANES and EXAFS regimes. Experiments were carried out in plants (rhizomes, roots and leaves) collected and laboratory cultivated *Imperata cylindrica* from Rio Tinto river (Spain). Predominant compounds of iron detected in rhizomes and roots correspond to jarosite and ferromagnetic iron oxides (ferrihydrite, hematite, and spinel phases), with less iron content in leaves. They converted to nanocrystalline maghemite after calcination process. These results have been summarized in the following published article: *Formation of biomineral iron oxides compounds in a Fe hyperaccumulator plant: Imperata cylindrica (L.) P. Beauv.* V. Fuente, L. Rufo, B. H. Juárez, N. Menéndez, M. García-Hernández, E. Salas-Colera, and A. Espinosa. *J. Struct. Biol.* 193, 23-32 (2016).¹

Results:

In Fig. 1, we present the XANES spectra at the XANES regime of experimental samples (roots and rhizomes) and the iron oxides compounds as references (jarosite, ferrihydrite, hematite, maghemite and magnetite) as possible candidates. The most intense transition observed in the absorption spectra is due to the allowed 1s to 4p electron transition. All spectra of *I. cylindrica* samples, including collected roots and rhizomes (fresh and calcinated) and cultivated in the laboratory exhibit an overall Fe absorption of Fe³⁺. No ferrous iron has been detected as reported other authors studying *Triticum*.² However, some differences are detected in the shape of absorption spectra: XAS results reveal that for the calcinated sample, the shape is well defined in contrast to either fresh collected and cultivated plants.

These differences can be attributed to crystallization occurred when the sample is subjected to a heating, mainly in form of maghemite. On the other hand, the spectra of fresh samples can be reproduced as a combination of contribution from ferrihydrite (60%), jarosite (20%) and a mixture of magnetite, hematite and maghemite (20%). Nanosize effects can also modify the bulk references spectra. In case of magnetite, its reference presents an absorption edge between Fe²⁺ and Fe³⁺, however, the overall valence state in measured plants is 3+. The samples cultivated in laboratory conditions would differ in the percent of combination of these oxides, which is in agreement with Mössbauer measurements that evidenced a decrease of jarosite content in comparison with fresh samples. These compounds would be reduced probably into crystalline maghemite when heated.

A further structural analysis has been carried out extracting the signal in the EXAFS regime after the absorption edge. Fig. 2a and b show the EXAFS signals and their respective Fourier transforms. Fourier transform magnitudes are related to coordination shells around the iron atoms. It can be observed that the first coordination peak is similar for all samples, which is corresponds to a distance Fe-O around 2.0 Å. This distance is in agreement with the mean radius Fe-O of iron lattice in a cubic and spinel structure corresponding to ferrihydrite, jarosite, maghemite, hematite and magnetite species, which ranges from 1.96 to 2.05 Å.⁴⁻⁷ However, their initial spectral features of samples resemble to the corresponding of the reference ferrihydrite⁷⁻⁸ characterized by rounded peaks at same positions.

Low intensity secondary coordination peaks are also detected at distances around 3–3.5 Å, which are also a characteristic of the above structures (with Fe–Fe distances between 3–3.65 Å and Fe–S at 3.22 Å), however, the amorphous nature of samples hamper the contribution of outer shells, diminishing the signal intensity signal.

On the other hand, spectrum feature of calcinated sample exhibits a well-defined chi structure and a small shoulder at 5 \AA^{-1} . Moreover, a second coordination peak that would be account to the Fe–Fe shell is observed, indicating an increase of number of neighbors. This could correspond to crystallization from an amorphous structure, mainly as maghemite.

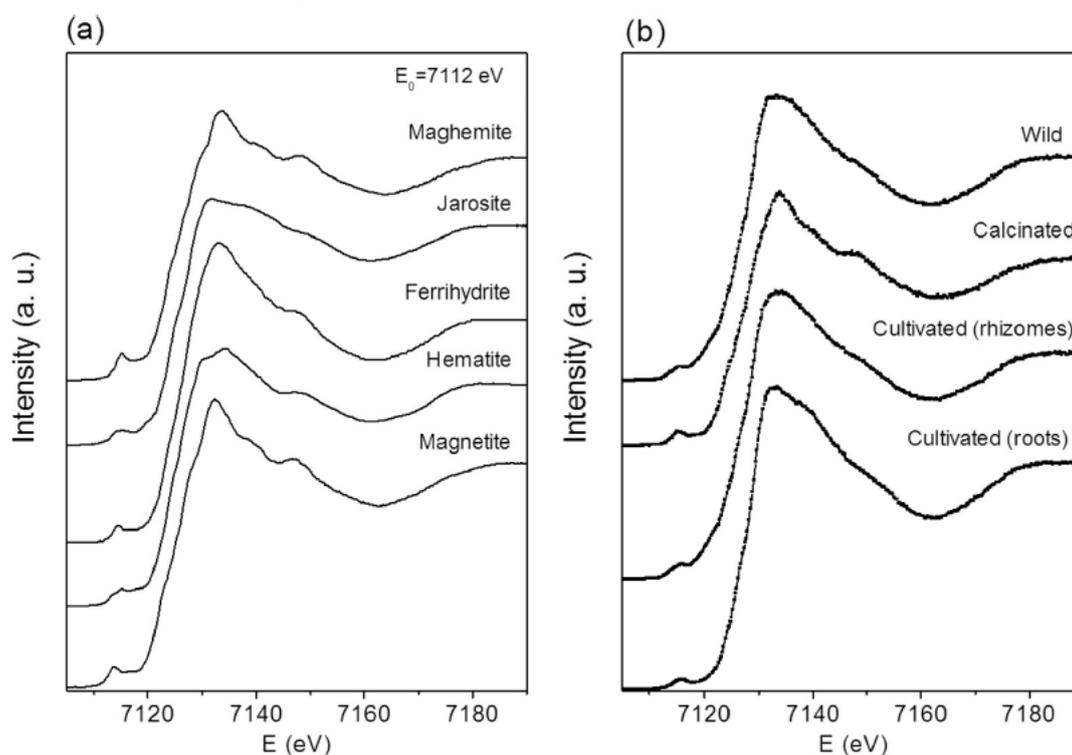


Fig. 1. XANES spectra at the Fe K-edge of (a) iron oxide reference compounds and (b) *I. cylindrica* samples (from roots and rhizomes).

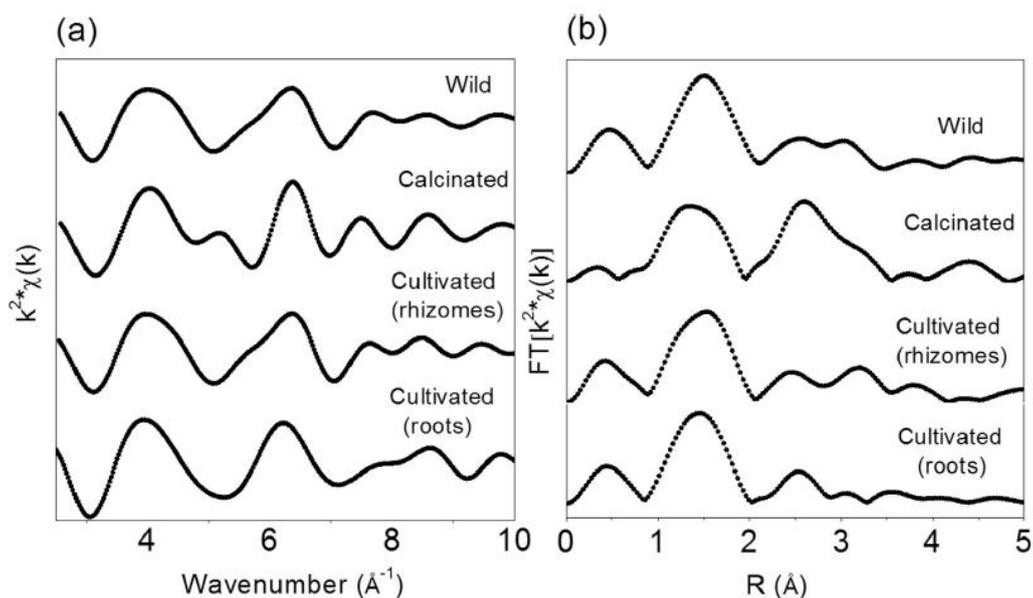


Fig. 2. $k^2 \cdot \chi(k)$ spectra at the Fe K-edge and Fourier transformed EXAFS spectra of the respective samples (roots and rhizomes).

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