



	Experiment title: Investigation of chemical form of iron in grain tissues of diverse wheat genotypes	Experiment number: LS- 2270
Beamline: ID21	Date of experiment: from: 7.11 to: 12.11.2013	Date of report:
Shifts: 15	Local contact(s): Hiram Castillo-Michel	<i>Received at ESRF:</i>
Names and affiliations of applicants (* indicates experimentalists): Katarina Vogel-Mikuš^{a,b*}, Peter Kump^{b*}, Iztok Arčon^{b,c*}, Sudhir Singh^d ^a Biotechnical faculty, Dept. of biology, Večna pot 111, SI-1000 Ljubljana, Slovenia ^b Jožef Stefan Institute, Jamova 39, POB 000, SI-1001 Ljubljana, Slovenia ^c University of Nova Gorica, Vipavska 13, POB 301, SI-5001, Nova Gorica, Slovenia ^d National Agri-Food Biotechnology Institute, Department of Biotechnology (DBT), C-127, Industrial Area, Phase VIII, Mohali 160071, India		

Report:

Iron is an essential element for all higher organisms assuming many important biological roles [1]. Although Fe is abundant in soils, this element is not readily accessible to the plants. According to the World Health Organization, Fe deficiency is the most common nutritional disorder in the world, affecting nearly 30% of the world's population. Delivery of iron via micronutrient rich bio-fortified cereal grains would offer a sustainable and food-based approach for addressing iron malnutrition. In order to develop Fe rich wheat cultivars, knowledge of basic mechanisms of iron uptake, transport and accumulation is needed [2].

Within this beamtime Fe localization, speciation and ligand environment of 4 wheat (IITR26 = high Fe, WL711 and WH291 = low Fe, *Aegilops kotschii* = high Fe), and 4 pearl millet genotypes with differential Fe uptake was studied. The beamtime was successful and all the proposed goals were accomplished.

High wheat Fe genotypes showed distinct accumulation in the aleurone layer, scutellum and in one genotype (*Aegilops kotschii*) in the pigment strand (Figure 1). Endosperm was highly depleted in Fe, therefore Fe-K XANES analysis was not possible.

Fe-K XANES (Figure 2) in wheat grains revealed tissue and genotype specific ligand environment (Figure 3). Best linear combination fit was for the all measured spectra obtained with Fe²⁺-phytate/ sulphate, Fe³⁺-phytate and Fe³⁺-citrate. In some spectra radiation damage was observed - shifting Fe-K edge towards lower energies and increasing the level of Fe²⁺. These spectra were discarded.

As shown in Figure 3 high Fe genotypes (IITR26 and A.kot) are showing higher percentage of Fe³⁺ phytate in aleurone than other low Fe genotypes indicating that Fe homesotaxis is highly regulated in grains and excess Fe is bound to phytate in high Fe genotypes.

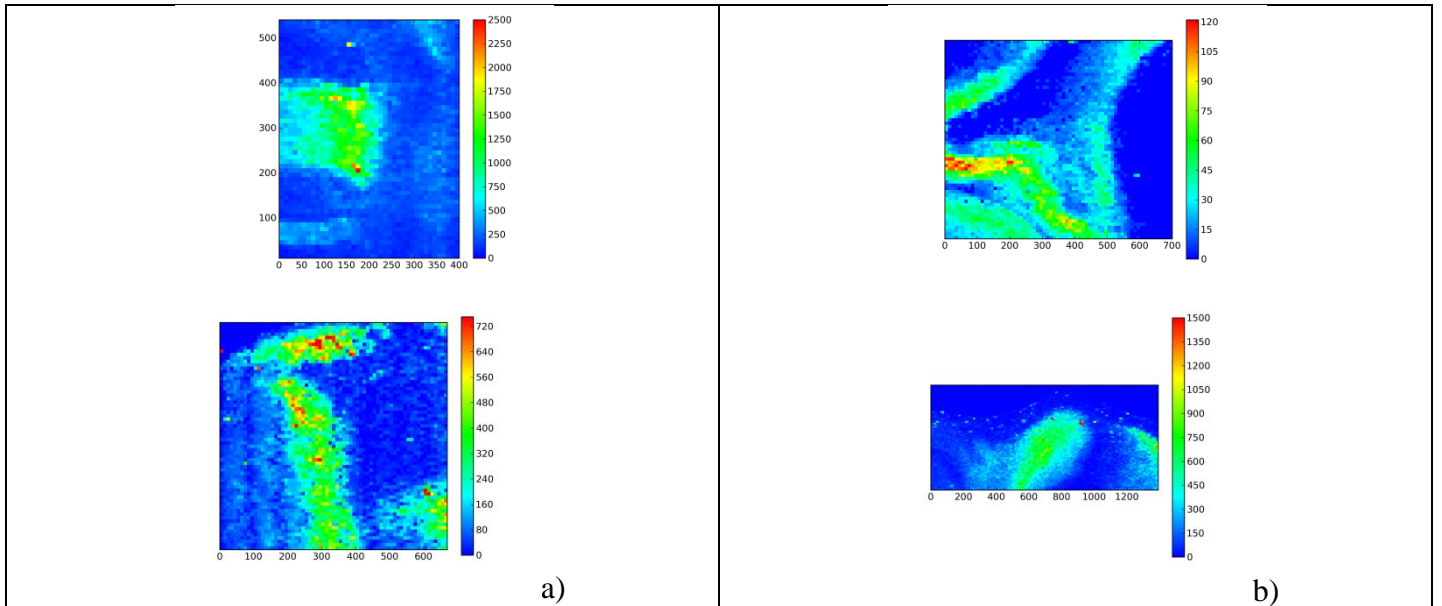


Figure 1. Quantitative images of Fe distribution of two wheat genotypes. A) *A. kot.* - pigment strand (upper) and scutellum (lower), b) WH291 - nucellar projection (upper) and scutellum (lower).

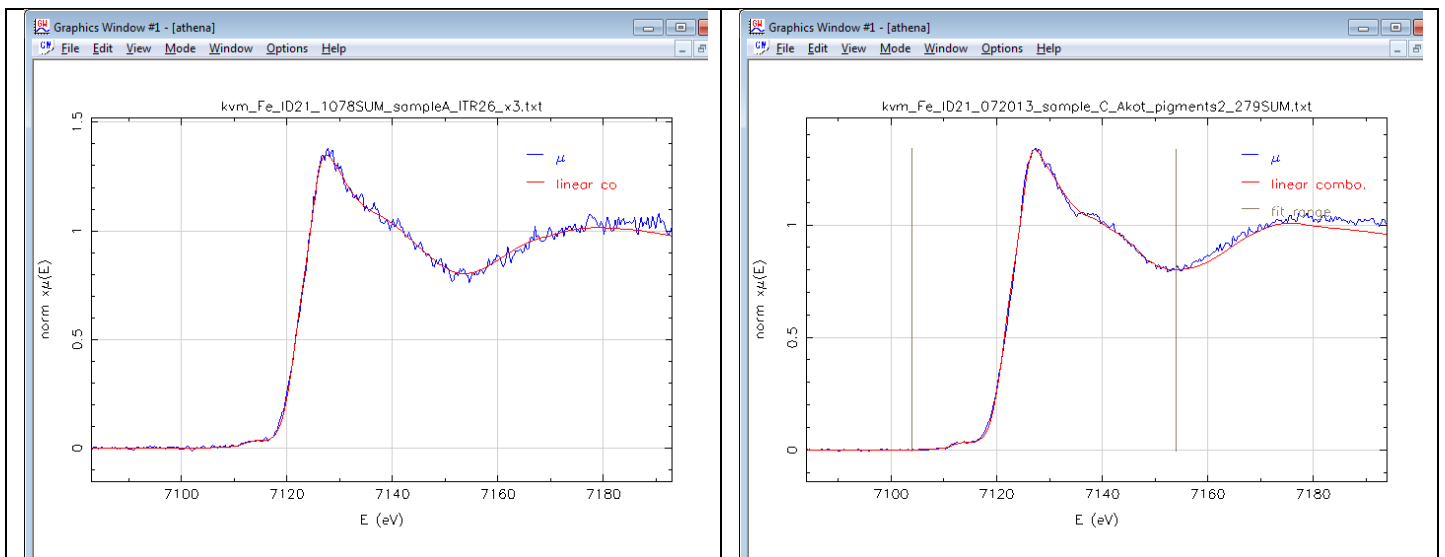


Figure 2. Fe-K XANES spectra recorded with focused beam in different tissues of wheat grain a) sample IITR26, aleurone, b) sample *A.kot*, pigment strand.

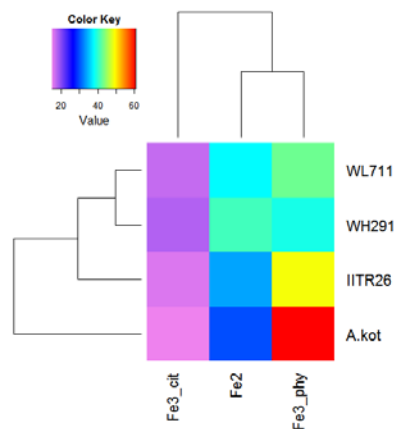


Figure 3. Genotype specific Fe ligand environment in aleurone of different genotypes showing the percentage of each ligand (Fe²⁺-phytate, Fe³⁺-phytate and Fe³⁺-citrate). Heatmap with cluster analysis was produced in "RGui" using gplots plug-in.