Zinc and Iron Speciation in the Barley Grain

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INTRODUCTION

Iron and Zn in the cereal grain are bound in molecular species (complexes), which reduce the bioavailability of these essential elements to humans and monogastric animals. In areas of the world where the human diet is primarily based on cereals, this leads to development of Fe- and Zn-deficiency related diseases. The most recent estimates indicate that approximately 50% of the global population suffer from Zn and Fe deficiencies, predominantly in third world countries.

The cyclic alcohol *myo*-Inositol is utilized in several biochemical pathways in plants, and has numerous phosphorylated derivatives of which phytate (*myo*-inositol-1,2,3,4,5,6-hexa*kis*phosphate; Ins P_6) is the most abundant one. Besides from being the major P compound, phytate is assumed to play an essential role in the storage of Fe and Zn in the cereal grain. However, it has not been confirmed that phytate alone is responsible for the low bioavailability of Fe and Zn in the cereal grain, and not much is known about the interaction of Fe and Zn with proteins and starch. This study was initiated to analyse these aspects.

METHODS

The speciation of Fe, Zn and P was studied using Liquid Chromatography - Inductively Coupled Plasma-Mass Spectrometry (LC-ICP-MS), which is a sensitive multi-elemental technique allowing analysis of how a wide range of elements are distributed between ligand fractions in the cereal grain. In this study, LC-ICP-MS was combined with Electrospray Ionization-Time Of Flight-Mass Spectrometry (ESI-TOF-MS) to obtain structural information about the molecular species.

RESULTS AND DISCUSSION

Multi-elemental analysis of various grain tissues of barley (*H. vulgare* cv. Golden Promise) confirmed that Fe and Zn were predominantly confined to the bran, embryo and aleuron layer whereas only marginal traces were found in the endosperm. Tissue staining using the Zn-specific stain (diphenylthiocarbazone) confirmed that Zn was accumulated in the alueron layer whereas no significant staining was observed for the endosperm (Fig. 1).

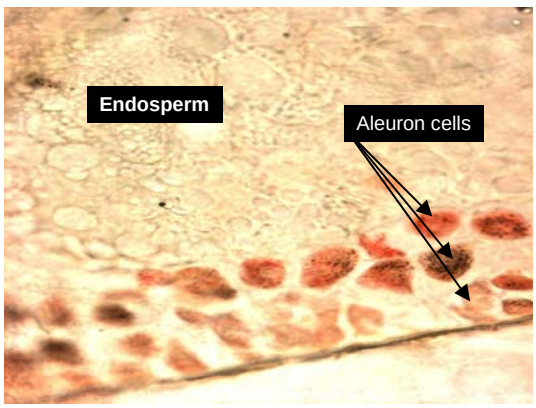

Fig. 1. Zinc specific staining of the barley grain showing that Zn is concentrated in the three cell layers of the aleuron tissue.

The LC-ICP-MS analysis identified seven different P species in the barley grain, one of them, namely Ins-P, was the dominating species. It is noteworthy that only three of these P-species appeared to bind Fe (Fig. 2). In contrast, only one Zn species was identified, and it was not associated with any P peak, showing that Fe and Zn speciate differently in the barley grain. Zinc appeared to be associated exclusively with a 7 kDa ligand, whereas Fe was bound to P in 5, 6 and 6.5 kDa species. These ligands are currently being analysed, and the results of these studies will be presented at the conference.

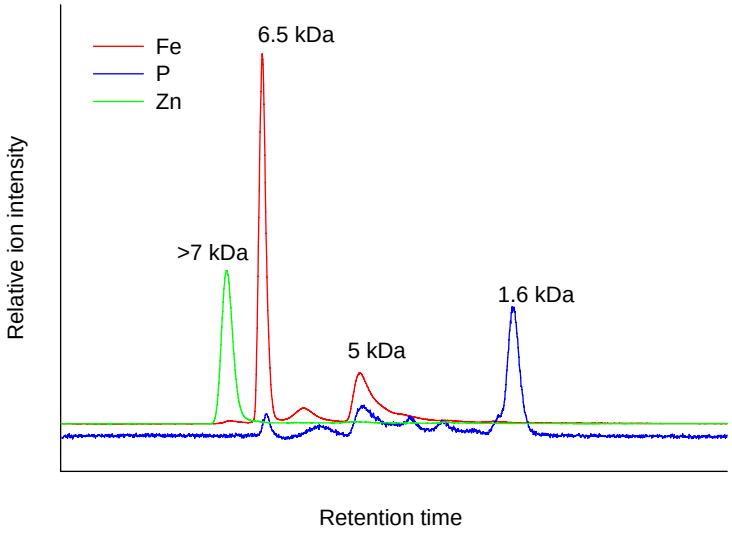

Fig. 2. Size exclusion chromatography hyphenated with ICP-MS identified a differential binding of Fe and Zn in the barley grain. Seven P species were fractionated, three of them were binding Fe. In contrast, Zn was not associated with P.

ACKNOWLEDGEMENTS

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