Zinc dependant polypeptides in different compartments of *Lathyrus- Rhizobium* sp. Symbiosomes

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SUMMARY

To analyze the effect of zinc deficient condition on N_2 -fixing root nodules, the proteins were analyzed in different compartments of *Lathyrus* – *Rhizobium* sp. Symbiosomes as a model symbiotic system. *Lathyrus sativus* plants were grown in pots containing *Rhizobium* – inoculated acid washed river sand irrigated by nitrogen free plant growth medium with and without zinc. Symbiosomes, isolated from N_2 - fixing root nodules were sub-fractionated and protein profiles in the different fractions (host plant cytoplasm, peribacteroid space and bacteroid cytosol) were analyzed through SDS-PAGE. In all different fraction high protein content was observed in plants grown at optimal zinc concentration of 1.0mg/L. The increase in protein was due both to a general increase in abundance of the most dominant polypeptides and to a pronounced increase in the abundance of specific polypeptides. Besides the unique pattern of protein in each compartment, polypeptides of same molecular size were also observed in the symbiotic interface PBS.

Key words : Lathyrus sativus - Rhizobium, Symbiosomes, Zinc, Polypeptides

Zinc has a unique place in the biology of planet earth. About 50% zinc deficient soil of the developed and developing world is recognized as a serious threat to both crop production and human health globally. Across all phyla from bacteria to humans, more proteins bind or require zinc for their function than those binding all other biologically essential cations combined (Gladshev *et al.*, 2004; Cakmak, 2008).

Zn is an essential catalytic component of over 300 enzymes and metalloenzyme complexes. It also plays a critical structural role in many proteins. For example, several motifs found in transcriptional regulatory proteins are stabilized by Zn, including the Zn finger, Zn cluster, and RING finger domains. Inside cells, Zn is neither oxidized nor reduced; thus, the essential roles of Zn in cells is based largely on its behavior as a divalent cation that has a strong tendency to form stable tetrahedral complexes. The Zn deficiency in plants has been recognized as a serious problem. Zn deficiency remarkably depresses the protein content and affects their polypeptides composition in the cells. Zn deficiency may be diagnosed by a combination of visual symptoms and soil analysis.

Despite the importance of Zn as an essential

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micronutrient for plant growth, it acts as environmental toxic factor and is known to affect the nodulation and dinitrogen fixation. The toxic effects of zinc and other heavy metals are explained by their interaction with sulphahydryl groups and concomitent inactivation of proteins (Assche and Clijesters, 1990). Therefore, it is generally accepted that tolerance towards zinc stress is based on mechanisms that maintain optimum and low free concentrations in the cytoplasm In addition, ressearchers have already suggested a role of zinc in maintaing the membrane integrity and H⁺ - ATPase dependent pH gradient across the membrane. It is essential to investigate the role of zinc in the symbiotic dinitrogen fixation process because of the importance of the relationship established across the membrane between the legume and Rhizobium.

Root nodulation starts with a molecular dialogue between two partners and takes place through a series of developmental stages. During nodule development the rhizobia are released from the infection thread into the host cortical cell as bacteroids in a process resembling endocytosis, forming a new compartment within the host cell, the symbiosome (Roth *et al.*, 1989).We still do not know the detail mechanisms underlying Zn dependency and N₂-fixation of grasspea. Therefore, present study was intented as an effort in this direction.

MATERIALS AND METHODS

Seeds of *Lathyrus sativus* (Grass pea) were procured from local farmers of Doon valley and germinated seedlings were used for nodule isolated