

Molecular physiology of nickel and cobalt homeostasis in *Rhizobium leguminosarum*.

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INTRODUCTION

Transition metals such as Fe, Cu, Mn, Ni, or Co are essential nutrients, as they are constitutive elements of a significant fraction of cell proteins. Such metals are present in the active site of many enzymes, and also participate as structural elements in different proteins. From a chemical point of view, metals have a defined order of affinity for binding, designated as the Irving-Williams series (Irving and Williams, 1948)



Since cells contain a high number of different proteins harbouring different metal ions, a simplistic model in which proteins are synthesized and metals imported into a “cytoplasmic soup” cannot explain the final product that we find in the cell. Instead we need to envisage a complex model in which specific ligands are present in definite amounts to leave the right amounts of available metals and protein binding sites, so specific pairs can bind appropriately. A critical control on the amount of ligands and metal present is exerted through specific metal-responsive regulators able to induce the synthesis of the right amount of ligands (essentially metal binding proteins), import and efflux proteins. These systems are adapted to establish the metal-protein equilibria compatible with the formation of the right metalloprotein complexes. Understanding this complex network of interactions is central to the understanding of metal metabolism for the synthesis of metalloenzymes, a key topic in the *Rhizobium*-legume symbiosis. In the case of the *Rhizobium leguminosarum* bv *viciae* (*Rlv*) UPM791 - *Pisum sativum* symbiotic system, the concentration of nickel in the plant nutrient solution is a limiting factor for hydrogenase expression, and provision of high amounts of this element to the plant nutrient solution is required to ensure optimal levels of enzyme synthesis (Brito *et al.*, 1994).

Usually, transition metals are present at low levels in natural habitats, and specific, high affinity transport systems have been developed by bacteria to import them from the soil solution. At the same time, these elements are toxic even at moderate concentrations, since they can catalyze the production of free radicals, or compete with other metals for the active site of metalloenzymes. In this talk we will present our recent work on the mechanisms involved in the homeostasis of transition metals nickel and cobalt in free living and symbiotic cells of the endosymbiotic bacterium *Rhizobium leguminosarum*.