ABSTRACT

This PhD thesis is based on a collaboration between Professor Dr. Ramon Serrano's laboratory and the fertilizer company Fertinagro Nutrientes and it's motivated by the increasing need in agriculture to increase crop productivity while minimizing its impact on the environment. Fertinagro Nutrientes supplied the following six bioestimulants for analyzing its effects on tolerance to abiotic stress in the yeast *Saccharomyces cerevisiae*, providing no (or very limited in some cases) information about its composition: Seaweed extract, Vinacillas, Phosphite, Humic Acid, Amino22 and Potassium fulvate. The broad conservation of metabolic pathways and the molecular mechanisms involved in abiotic stress response between yeast and plants allows the use of *S. cerevisiae* as a suitable model system for this study.

All bioestimulants showed, to a greater or lesser extent, positive effects on yeast tolerance against at least one of the abiotic stresses in study. Seaweed extract improves growth under salt stress. Phosphite and Vinacillas protect against salt stress and heat shock. Humic acid increases tolerance to oxidative stress and heat shock. The two most versatile bioestimulants, Amino22 and Potassium fulvate, were further investigated in order to identify their mechanisms of action.

Amino22, which is the most effective biostimulant, considerably improves growth in the absence of stress and under osmotic stress and, to a lesser extent, it also enhances growth under salt stress conditions. Amino22 also provides tolerance against oxidative stress and heat shock. By comparison with an acid casein peptone it was confirmed that the active substance in Amino22 are amino acids. Analysis of global gene expression using DNA microarrays showed that Amino22 treatment represses both Gcn4 regulated genes (which are involved in amino acid and vitamin biosynthesis) and Aft1 regulated genes (which are involved in iron homeostasis). The positive effect of amino acids on growth and stress tolerance is related, at least in part, to the activation of TORC1 pathway and it also requires a partial inhibition of GAAC pathway. By contrast, this positive effect is independent of the Aft1 regulon repression. Following the observation that amino acid treatment represses iron regulon expression, we studied more in depth the relationship between amino acid and iron homeostasis in *S. cerevisiae.* We suggest a regulation model according to which the activation of GAAC pathway induces nuclear localization of Aft1 and subsequent expression of its target genes, whereas inhibition of this pathway by amino acids may have the opposite effect. Aft1 activity may be controlled through eIF2α phosphorylation and, hypothetically, it would involve regulation Fe/S biosynthesis.

Potassium fulvate improves tolerance to osmotic, oxidative and heat stress. Microarray and qRT-PCR expression analysis indicate that Potassium fulvate represses Aft1 regulon. This repression can be explained by an increase in intracellular iron content, whose absorption depends on an oxidoreductase encoded by the *FET3* gene. Tolerance to abiotic stress by Potassium fulvate also depends on *FET3*, therefore it is concluded that the mechanism of action of this biostimulant is based on an increased iron availability, which is accumulated in yeast cells without causing oxidative damage.