Abstract:

The GCN2 protein kinase is a conserved protein in all eukaryotes involved in translation control under stress conditions. It is considered a key point in the control of cellular homeostasis and a sensor for a wide variety of stress conditions. Aminoacid fasting was the stress that started its characterization in yeast and animal, but recently activation of this system has been observed in both biotic and abiotic stresses. The GCN system has been extensively described in Saccharomyces cerevisiae: GCN2 binds to GCN1 and GCN20 proteins, allowing kinase activation in aminoacid fasting situations. GCN2 is activated by uncharged tRNAs, and subsequently phosphorylates the translation factor eIF2α, leading to a reduction in overall protein synthesis, but also a greater translation of specific mRNAs, such as those encoding GCN4. This transcription factor will regulate the expression of new genes, allowing the cell to initiate an adaptive response to stress.

In plants, it is not deeply known how the GCN system helps to alleviate stress and control homeostasis. All three known proteins in this system have homologs in *Arabidopsis*. Some studies indicate that the mechanism of action of GCN2 in plants presents many gaps. While plant GCN2 kinase is activated under different stress situations, the involvement of GCN1 and GCN20 homologs in these processes is controversial, and recently it has been proposed a new role for GCN1 in translation, independent from GCN2.

The GCN1 homolog in plants is involved in innate and acquired immunity and its mutant lines present very different phenotypes from those of the GCN2 mutant lines. The functional relationship between these two genes is difficult to define in plants. In this thesis, we prove that, although the *Arabidopsis* GCN1 and GCN2 genes are necessary to mediate in the phosphorylation of eIF2α after treatments with glyphosate, an inhibitor of aromatic aminoacid biosynthesis, the loss of function mutants of both lines develop different phenotypes of root and chloroplast. Electron microscopy experiments reveal that the mutants in GCN1, but not in GCN2, are affected in chloroplast biogenesis, which explains the macroscopic phenotype previously observed for these mutants. The mutants in GCN1 present a complex transcriptional reprogramming that affects, among others, the responses related to defense mechanisms, photosynthesis and the correct folding of proteins. Analysis of the double mutants suggests that GCN1 in plants has another function, which is independent from phosphorylation of GCN2 and eIF2α. These results show that both genes have common and different functions in *Arabidopsis*.

On the other hand, we show that none of the five GCN20 homologous genes in *Arabidopsis* is necessary for the phosphorylation of eIF2α. Furthermore, the phenotypes under abiotic stress of mutant plants in them, and the development of their chloroplasts, suggest that GCN20 is functionally related to GCN1, but not to GCN2, which is confirmed because the *gcn1* and *gcn20* mutants share a similar transcriptional reprogramming and affects photosynthesis and stress responses.

We identify the GCN2 protein kinase as a cellular component that promotes the action of glyphosate in *Arabidopsis*. Comparative studies using GCN2 loss-of-function mutant seedlings show that the molecular program that the plant develops after the treatment with the herbicide is not taking place. Furthermore, adult *gcn2* plants show less inhibition of photosynthesis, and accumulate less shikimic acid than wild-type ones after glyphosate treatment. Something similar happens after treatment with UV-B ultraviolet light, where loss-of-function mutants are more resistant. Activation of GCN2 in the face of this stress is independent of the UV-B photoreceptor (UVR8) and its downstream signaling components and the stress signaling pathway of MAP kinases.