Degradation of autophagic bodies in sugar-starved lupin embryo axes: a transcriptomic and proteomic approach

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The degradation of autophagic bodies is one of the final stages of autophagy. Autophagic bodies are spherical structures formed in the vacuole of yeasts and plants. The newly formed autophagic bodies are rapidly degraded by vacuolar lytic enzymes. Based on our results, autophagy is significantly enhanced by sugar starvation in cells of lupin embryo axes but asparagine (a central amino acid in lupin seed metabolism) causes a clear inhibition of autophagic bodies degradation. Trying to describe the role of asparagine in a mechanism of autophagic body degradation, we performed transcriptomic and proteomic analyses of lupin embryo axes. The experiments were performed on embryo axes isolated from imbibed seeds of white and Andean lupin. Embryo axes were cultured in vitro for 96 h on a mineral medium supplemented with 60 mM sucrose, without the sugar, and on both the media mentioned above enriched in 35 mM asparagine. The quality of the libraries was verified by Sanger sequencing method, and the large-scale transcriptomic sequencing using Illumina HiSeq Next Generation Sequencing technology (NGS) was performed. The obtained sequence reads were aligned to reference transcriptome and counted in the aim to find differentially expressed genes. As transcriptome modulation could be manifested in proteomic changes, isobaric tags for relative and absolute quantitation (iTRAQ)-based proteomics was performed to screen the differentially expressed proteins. Our goal was to analyze the effect of asparagine on the expression of genes and accumulations of proteins involved in autophagy. First of all, we focused on changes in the level of transcripts of genes coding for ATG proteins and genes coding for vacuolar lytic enzymes (e.g., proteases) as well as on the changes in the accumulation of appropriate proteins.

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