Biochemical Characterization of Interactions of the Yeast Exosome with Regulatory Factors

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The exosome is a protein complex formed by eleven subunits, which is present in archaea and eukaryotes, has exoribonucleolytic activity in the 3'-5' direction, and is involved in processing and/or degradation of all kinds of RNA. Our laboratory has been working on the functional and structural characterization of the archaeal and yeast exosomes, obtaining a model for the general structure of the yeast complex based on subunits interaction data. Exosome complex is involved in different pathways of RNA processing and degradation, and because it interacts with single-stranded RNA in a non-sequence specific manner, it is currently predicted that the control of exosome activity is done by proteins that interact both with RNA and the exosome. This work aims at the functional and structural characterization of the role these regulatory proteins play in the control of the exosome activity and the effects of these interactions on the complex structure. We started the purification of the yeast exosome complex by using the the TAP-tag (Tandem Affinity Purification) technique, followed by the identification of proteins co-purifying with the complex by mass spectrometry. The nuclear exosome was purified with subunit TAP-Rrp6p and the identification of interacting partners revealed some exosome subunits and also new potential exosome regulatory proteins, which play an important role in the control of posttranscriptional control of gene expression.

Keywords: Exosome, TAP-tag, yeast, RNA

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