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Biochemical identification and analysis of ADT-like proteins from plants

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Phenylalanine (Phe) biosynthesis in plants is a fundamental process as Phe serves as a precursor of proteins but also of a massive array of essential phenylpropanoids for plant growth development defense or reproduction. Arogenate dehydratase (ADT) catalyzes the final and rate limiting step in the Phe biosynthetic pathway in plants the conversion of arogenate into Phe. In recent years different studies have shown that in most plant species multiple isogenes encode for alternative ADT isoenzymes that display partially overlapping roles in different aspects such as lignin accumulation or the size and mass of the stems (Corea et al. 2012a Corea et al. 2012b) but also display exclusive functions in processes related to development (EI-Azaz et al. 2018) or accumulation of certain metabolites (Chen et al. 2016). Very interestingly in recent years it has been determined that some ADT isoenzymes also display prephenate dehydratase activity (PDT) and thus are able to participate in an alternative route of Phe synthesis in plants using phenylpyruvate as intermediary (EI-Azaz et al. 2016).

In our article EI-Azaz et al. 2016 and using phylogenetic studies that included ADTs from multiple plant species corresponding to most plant clades we identified several sequences that encoded for proteins with partial similarity to plant ADTs that we have provisionally named as ADT-like proteins. Prominently in conifers we identified up to four ADT-like enzymes. Using functional complementation assays in yeast we have determined that these enzymes do not display PDT activity but remains unclear its putative ADT or alternative activity. Using biochemical structural and functional analysis we have investigated the role of these enzymes in the *Pinus pinaster* conifer species. This communication will discuss on how the neo-functionalization of these enzymes may be related to the existing metabolic specialization in some groups of plants.

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