

# TANnase Tools for Renewable Aromatic building blocks

## OBJECTIFS

The general approach is to find new alternatives to the overused phenolic chemicals in order to i) substitute hazardous specialties by safer & natural ones while avoiding the use of petrochemicals, ii) valorize agricultural and forest co-products and wastes while expanding local economies, iii) diversify and develop original compounds for existing or new applications. This study targets tannins, which are the second most abundant source of natural polyphenols. However, these biopolymers require to be depolymerized into small phenolic molecules, that may be further functionalized, before their use as fine chemicals. For this, the development of a biocatalyst toolbox specific to tannins would be highly valuable. The project aims at identifying and characterizing new tannases.

## RESULTATS

Structural analogs of the natural substrates of tannases were synthesized in order to identify chemical patterns involved in tannase affinity and their catalytic activity. Their application for the study of commercially available tannases showed that the hydrolysis of complex glucogallins and flavanoyl-3-O-gallate, was significantly faster than that of simple alkyl gallates. New microbial tannases were produced by fermentation on solid vegetal substrates. The highest enzyme productions were obtained on industrial fruit co-products, especially on pomegranate peels. This non-gmo approach generates complex enzyme extracts but is the most suitable for food applications. In order to allow the selective purification of tannases, affinity supports with grafted structural analogs of selected target substrates were designed. This approach was completed with the construction of recombinant strains to allow the heterologous production and the protein engineering of selective biocatalysts for fine chemistry.

## PERSPECTIVES

Tannases are currently mainly used in the food industry to decrease astringency and bitterness of beverages. The production and the fine characterization of an extended set of tannases will be a valuable contribution to the enzymatic toolbox available for the molecular engineering of natural polyphenols for research and industrial applications.

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