PAPAYA LIPASES HETEROLOGOUS EXPRESSION: TOWARD STRUCTURE AND FUNCTION RELATIONSHIP

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Over the last years, plants have been studied as a source of new lipases with interesting properties for industrial applications. Only one esterase (CpEst) has been extracted directly from *Carica papaya* latex [1], mainly because it is extremely difficult to purify these enzymes from latex due to their high attachment to the insoluble fraction. To overcome this problem, heterologous expression of these proteins presents a suitable alternative to obtain higher quantities of protein. In addition, in some cases catalytic properties of these lipolytic enzymes have not been reported yet and therefore it is interesting to evaluate their expression kinetics, function, biochemical properties and possible applications. In fact, *Carica papaya* latex proteins, have a large variety of applications such as synthesis of biopolymers [2], human fat milk analogs [3], medium and long chain diesters of 2-oxoglutaric acid [4] and enantiomeric resolution [5]. Cloning and expression of several *C. papaya* lipases were performed in *Pichia pastoris*. Lipase activity was measured spectrophotometrically using p-nitrophenyl butyrate as substrate or by titration of olive oil hydrolysis [6]. Three-dimensional structure modeling was also performed (Figure 1). Our methodologies allowed cloning of several *C. papaya lipases* in *Pichia pastoris*. RhB/Tributyrin plate screening allow identification of clones from CpLip1, CpLip3 and CpEst that present lipase activity. Further efforts on papaya lipases expression should lead to identify the proteins responsible of the activity observed in papaya latex and to preparations with higher specific activity.

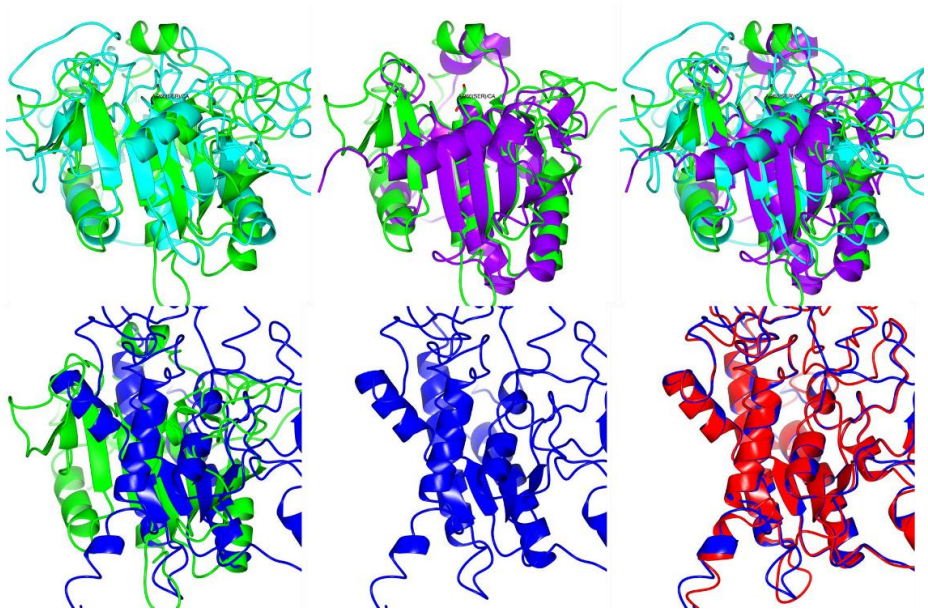

Figure 1 - Protein three-dimensional structure model of CpLi1 ■, CpLip3 ■, CpLip5 ■, CpLip6 ■ and CpEst ■; in different superposition.

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