

Epoxide hydrolase from the fungus *Trichoderma reesei*: biochemical properties and conformational characterization

Oliveira, G.S.¹; Adriani, P.P.¹; Lopes, A.R.²; Campana, P.T.¹; Chambergo, F.S.¹

¹Escola de Artes, Ciências e Humanidades, Universidade de São Paulo, São Paulo, Brazil; ²Instituto Butantan, São Paulo, Brazil.

INTRODUCTION: Epoxide hydrolases (EH; EC 3.3.2.3), are enzymes that catalyze the conversion of epoxides into their corresponding diols through the addition of a water molecule. EHs have biotechnological potential in chiral chemistry, for the preparation of enantiopure epoxides, vicinal diols or fine chemicals. **OBJECTIVES:** In the present work, we report the cloning, purification, the enzymatic activity, and the conformational analysis by means of Circular Dichroism spectroscopy, of the TrEH gene from Trichoderma reesei strain QM9414. **MATERIALS AND METHODS:** The EH gene has an open reading frame encoding a protein of 343 amino acid residues resulting in a molecular mass of 38.2 kDa. The TrEH was purified and characterized using racemic styrene oxide (SO) as substrate. **DISCUSSION AND RESULTS**: The enzyme presents a pH optimum = 7.2, it is highly active at temperature range from 23 to 50 °C and thermal inactivation at 70 °C (t1/2= 7.4 min). The Michaelis constant (K_m) were 4.6 mM for racemic substrate, 21.7 mM for (R)-(+)-styrene oxide and 3.0 mM for (S)-(-)-styrene oxide. k_{cat}/K_m analysis indicated that TrEH is enantioselective hydrolyzing preferentially (S)-(-)-styrene oxide. The conformational stability studies suggested that, despite the extreme conditions (high temperatures and extremely acid and basic pHs). CONCLUSION: The recombinant protein showed an enantioselectivity distinct from that presented by other fungus EHs, making this protein a potential biotechnological tool.

Keywords: *Trichoderma reesei*, Epoxide hydrolase, Enantioselectivity Supported by: FAPESP 2015/03329-9 and CAPES