Purification and Partial Characterization of a Lipase from the Thermophilic Mold *Malbranchea pulchella*.

Pereira, M.G.; Guimarães, L.H.S.; Polizeli, M.L.T.M.; Terenzi, H.F.; Jorge, J.A.
Departamento de Biologia, Faculdade de Filosofia Ciências e Letras de Ribeirão Preto - USP, Ribeirão Preto, SP.

Lipases hydrolyze triacylglicerols to fatty acids and glycerol, and represent biocatalysts of choice for several industrial applications due to their unique chemo-, regio- and enatioselectivities, which enable the production of new drugs, agrochemicals and fine chemicals. Here we report the purification and partial biochemical characterization of a lipase from *Malbranchea pulchella* var. sulfurea. Lipase activity was produced in semi-solid cultures with wheat bran, and maximal enzyme production was achieved after 192 h growth at 40ºC. The lipase was purified to homogeneity by ammonium sulfate precipitation followed by DEAE-cellulose and Octyl-Sepharose column chromatography. The enzyme was purified about 41-fold and a specific activity of 0.42 U/mg protein was achieved. The purified lipase showed a single protein band both in 7% PAGE and 8% SDS-PAGE. The apparent molecular mass estimated from SDS-PAGE was 30 kDa. Optimum of temperature and pH for lipase activity corresponded to 50-55ºC and 7.0, respectively. The purified enzyme was completely stable in aqueous solution at 65ºC up to 1 h, and presented a half-life of about 37 min at 70ºC. Moreover, it was stable for 1 h when submitted to different pH values, ranging from 2.5 to 9.0. Metals ions had no effect on enzymatic activity. The purified enzyme exhibited apparent Km and Vmax values of 0.31 mM and 0.64 U/mg of protein, respectively, using p-nitrophenyl palmitate as substrate. The resistance of *Malbranchea* lipase to elevated temperatures as well as its stability in a wide pH range suggests that this enzyme has potential for biotechnological applications.
FAPESP, CNPq, CAPES.