Digestive α-fucosidase From *Tityus serrulatus* <u>Moreti, R.</u>¹, Lopes, A.R.¹ ¹Laboratório de Bioquímica e Biofísica, Instituto Butantan, São Paulo, Brazil

Tityus serrulatus females present α -amylase, chitinase, α -fucosidase, β glucosaminidase as the main digestive carbohydrases. α -fucosidases (EC 3.2.1.51) are glycosyl-hydrolases (GH, family 29), which catalyses the hydrolysis of glycosidic linkages (alpha 1,2, alfa 1,3, alpha 1,4 e alpha 1,6) present in glycoconjugates (carbohydrates, glycoproteins, glycolipids and glycosaminoglycans) between a fucose and other molecules. Few data on these enzymes are available to Arthropoda species. Hepatopancreas from Tityus serrulatus females were isolated and homogenized in Milli Q water in a Potter-Elvehjem homogenizer. α -fucosidase activity was measured using 0.05 mM MU α Fuc (4-methyl-umbelliferyl- α -L-fucoside) as substrate. Enzyme separation on gel filtration and anion-exchange chromatographies indicated that *Tityus serrulatus* presents two true α -fucosidases. This was shown by the assay of chromatographic fractions using α -glucosidase substrate (MU α Glu) and α -fucosidase substrates (MU α Fuc). The activities were found in different fractions indicating two distinct enzymes. These results were confirmed by heat inactivation using MU α Glu and MU α Fuc as substrates. α -glucosidase activity was completely inactivated at 65 °C while α -fucosidase remains active. The two α -fucosidase activities were also evidenced on isoeletric focusing, heat inactivation and in cation exchange chromatographies (Resource S column). Major digestive α -fucosidases from Tityus serrulatus was isolated by a combination of two cation-exchange chromatographies using a Hitrap S column and a Resource S column in a FPLC system. The major α -fucosidase activity was purified to homogeneity as demonstrated by a 12% polyacrylamide gel electrophoresis. These proteins presents a molecular mass of 86.5 kDa. The pH optimum of a-fucosidase was 5.0, the isoeletric point is 6.0. This α -fucosidase is not inactivated by temperatures of 50°C for 120 minutes.

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