

ROLE OF RESIDUE E190 IN MODULATING AGLYCON SPECIFICITY OF A β -GLYCOSIDASE FROM SPODOPTERA FRUGIPERDA (Sf β gli50-AF 052729) OF FAMILY 1.

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β -glycosidases are active upon a large range of substrates in part due to the region of the active site that interacts with the substrate that is formed by several subsites (1 to 3). This work has the objective to understand the role of residue into the interaction with the aglycone in a Sf β gli50. Site directed mutagenesis was used to replace this residue by an alanine. After this the mutant E190A was produced as recombinant protein in *E. coli* and purified by a hydrophobic chromatography and a ion exchange and characterized by measuring of K_m and k_{cat} for five p-nitrophenyl- β -glucosides and K_i for oligocellodextrins and alkyl- β -glucosides. Through of ΔG° for alkyl- β -glucosides binding was noticed that affinity for alkyl groups was not changed. But through of ΔG° oligocellodextrins binding it was noticed that the glucose affinity of subsites +1 and +3 decreased, but for the subsite +2 it was increased. Comparison of k_{cat} / K_m between mutant E190A and wild-type showed few changes in the interaction with the glycone. These results indicate that this residue has a small influence in the glycone binding and is probably localized in the subsite +1.

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