EFFICIENCY OF SHUFFED ALPHA-AMYLASE INHIBITOR IN INHIBIT *ANTHONOMUS GRANDIS* ALPHA-AMYLASE

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Anthonomus grandis is an insect that produces severe damage on cultivated cotton (*Gossipium hirsutum*). The induction of amylase synthesis in the digestive tracts of cotton boll weevil larvae and adults was correlated with the identification of starch accumulation in structures preferred by this pest for feeding and oviposition. The discovery and utilization of alpha-amylase inhibitors represents an efficient strategy to control boll weevils. However, alpha-amylase inhibitors including lectin class (αAI-1 and αAI-2) were unable to inhibit *A. grandis* alpha-amylases (AGA). Thus we developed strategies to create novel specificity in alpha-amylase inhibitors. A library of phage-expressed containing 10^7 alpha-amylase inhibitor variants produced by DNA shuffling of *αAI-1 and αAI-2* genes was screened against AGA. 5 out of 22 variants presenting affinity for AGA were introduced into *Arabidopsis thaliana* plants. The proteins from transformed plants were used to evaluate in vitro inhibition of AGA. Two mutants, showing activity higher than 70%, were purified and used for biochemical characterization as well as bioassays. The results have encouraged us to consider these variants to be used in constructions containing other genes (pyramidal effect). These vectors have been used to transform cotton plants to confer resistance to A. *grandis*.

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