

CHITINOLITIC ACTIVITY IN PROTEIC EXTRACTS OF *Bacillus thuringiensis*
TOXIC TO BOLL WEEVIL (*Anthonomus grandis*)

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Bacillus thuringiensis (*Bt*) is a spore forming bacteria, which produces Cry proteins toxic towards several insect orders. *Bt* S811 strain produces at least three Cry toxins: Cry1Ab, Cry1Ia12, and Cry8, and shown toxicity to insects from Coleoptera order. In order to characterize the production of these toxins, and check its activity against Boll weevil larvae, proteic extracts from *Bt* cells and supernatant proteins from the bacterial culture, were obtained at different stages of cell cycle; 8, 16, 24, and 32 hours after inoculation (HAI). Proteins from 32 HAI of the supernatant, and 8 HAI of the cellular fractions, shown highest activity towards the Boll weevil larvae. Western blotting assays using anti-Cry8 and anti-Cry1I were carried out to analyse these toxins in the *Bt* proteic extracts. The existence of a Cry8 was detected at 8 HAI in the cellular fraction, what allow associate this molecule with the toxicity of this fraction. However, toxicity observed at 32 HAI in the supernatant fraction, was not possible to be associated with Cry8 or Cry1Ia toxins, indicating that there are another protein(s) responsible for the toxicity. A protein homolog to Cry1Ab was identified by "Peptide Mass Fingerprint" at 32 HAI of the supernatant fraction and a chitin binding protein was identified by 2DE/MS/MS in this same stage and chitinolytic activity was also observed by enzymatic assay. All our data suggest a possible synergism between Cry toxins and a chitinase in the activity of this strain towards Boll weevil.

Keywords: Cry toxins, *Bacillus thuringiensis*, *Anthonomus grandis*, Chitinase
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