Structural Analyses of Cry 1Ac Protein from *Bacillus thuringiensis*

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ABSTRACT

Cry protein is a delta endotoxin of the Bacillus family that provides an entomopatogenic activity to *Bacillus thuringiensis*. These proteins have a specific toxic activity against three types of insect larva: Lepdopthera, Diptera and Coleopthera.

The insecticidal toxins are produced during spore formation. When an insect ingests these proteins they are activated by proteolytic cleavage. The toxin, after the ingestion, is solubilized by the alcalin pH in the digestive tract of the target insect. Once activated the endotoxin binds to the gut epithelium and causes cell lysis leading to death. These proteins are the active agents used in the majority of biorational pesticides and insect-resistant transgenic crops.

This activated region of the delta endotoxin is composed of three structural domains. Domain I is involved in membrane insertion, pore formation and toxicity. The second and third domains are involved in receptor binding and specifically domain III is important in insect specificity.

There are around 120 sequences of Cry toxins, and only five structures were deposited on the Protein Data Bank (PDB). There is a large interest in the toxin Cry 1Ac because it is commonly used to create transgenic plants with insect resistance.

A theoretical model of the Cry1Ac toxin was obtained on the basis of the coordinates of the insecticidal protein Cry1Aa (PDB code:1ciy.pdb) [1] as a template. The high sequence identity (73%) and a good correlation coefficient obtained from the elctron density server [2] indicates that 1ciy structure could be used as a template.. The model corresponds to residues 33-618 of the primary structure and consists of domains I, II and III. The sequence of domain I is highly conserved, while domain II and III have the lower sequence similarity. The 3D model was construct using Modeller v7.7. [3].

Aimed at verifying whether the amino acid differences in domains II and III could be responsible for insect specificity, this two structures were structurally aligned and compared with the Sting protein structure analysis program. Knowing that each amino acid substitution could potentially affect the protein structure and function, these amino acids were mapped and evaluated according to: conservation, change in solvent accessibility, side-chain volume change, effect on amino-acid interactions, protein electrostatics, and physicochemical properties of amino acids.

Such approach can open a new insight into understanding of specificity that Cry protein has for receptors in plant pests and can help in the design of mutagenesis experiments aimed to elucidate the mechanism of action of the Cry1Ac toxin.

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