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Isolation and SDS-PAGE analysis of dipteran toxic δ-endotoxin protein crystals of Sri Lankan *Bacillus thuringiensis* strains

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Bacillus thuringiensis (*Bt*) is characterized by its ability to produce parasporal, crystalline proteins known as δ-endotoxins. Several types of δ-endotoxins exhibiting different insecticidal activities have been identified including dipteran toxic *cry* genes 4, 10, 11 etc. Also a cytolytic protein, cytolysin (*cyt*1 and *cyt2*), is found in the crystal inclusions of the dipteran active strains. *Bacillus thuringiensis var. israelensis (Bti),* is toxic to dipteran larvae and known to contain *cry*4, *cry*10, *cry*11, *cyt*1 and *cyt*2 genes. The expression of these genes produces four proteins of 27 kDa, 65 kDa, 125 kDa and a doublet of 130 kDa. The objective of this work was to isolate and designate insecticidal crystal proteins in Sri Lankan *Bt* strains which has shown insecticidal activity against mosquito larvae of *Aedes aegypti* and *Culex quinquefasciatus* in previous studies.

In previous *cry* gene analyses, it was found that *Bt* isolates, AB58, AB107, AB110 and AB115 contained only *cry*4 and *cry*10 genes whereas isolates AB59, AB63, AB67, AB125, AB142, AB155, AB160, AB165 and AB172 contained *cry*4, *cry*10 and *cry*11 genes. *Cyt*1 gene was only found in *Bt* isolates AB67 and AB125. In comparison to *Bti*, only two local *Bt* isolates, AB67 and AB125 contain protein bands corresponding to 130 kDa, 67 kDa and 27 kDa and a light band at 27 kDa, supports the expression of *cyt*1 gene in these 2 isolates. *Bt* isolates, AB59, AB63, AB67, AB125, AB142, AB155, AB160, AB165 and AB172 contain protein bands corresponding to 130 kDa and 67 kDa and well defined band at 67 kDa suggests the presence of *cry*11 gene in these isolates. All the *Bt* isolates showed a protein band at 130 kDa confirming the presence of *cry*4 gene. Protein band at 78 kDa corresponding to *cry*10 gene was detected as a low intense band in SDS-PAGE analysis of *Bti*. Endotoxin proteins present in the crystals of the local *Bt* isolates were almost similar to *Bti*. This analysis further supports strongly the presence of the *cry* type of genes of local *Bt* isolates as in *Bti*. Further it can be concluded that expression of the *cry* and *cyt* genes of *Bt* are at a fairly good level under laboratory conditions.

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