

SUMMARY

The aim of present study was screening the soil samples from the local environment for a toxic *cry1* positive *Bacillus thuringiensis* (*Bt*). A total of 38 *Bt* strains were isolated, from different National parks, wild life sanctuaries, alluvial soil, leaf litter, insect habitat, stored product material, leaf surfaces of broad leaf trees and grain dust. All of these were characterized on morphological and biochemical basis. From these 5 isolates were found positive for *cry1* genes. The toxicity bioassays with *Bt* spores showed that six *Bt* isolates harboring *cry1* genes (*viz.*, GCU-*Bt* 1.5/1, GCU-*Bt* 3.1/1, GCU-*Bt* 6.1/1, GCU-*Bt* 11.1/2, GCU-*Bt* 14.1/1, and GCU-*Bt* 25.1/1) were most toxic to neonate larvae of lepidoptera, *Helicoverpa armigera*.

The 16S rDNA study revealed that these isolates showed maximum homology with *Bt* strains. The sequence alignment of 16S rDNA gene from GCU- *Bt* 1.5/1, 3.1/1, 11.1/2, 14.1/1 and 25.1/1, were submitted to GenBank database. GCU-*Bt* 1.5/1 showed 99% homology with *Bacillus thuringiensis* strain RX-MKV8 16S ribosomal RNA gene. GCU-*Bt* 3.1/1 showed 99% homology with *Bacillus thuringiensis* strain 2002007400 16S ribosomal RNA gene, partial sequence. GCU-*Bt* 11.1/2 showed 99% homology with *Bacillus thuringiensis* 16S rRNA gene and 16S-23S IGS, strain CMBL BT-1, While GCU-*Bt* 14.1/1 showed 99% homology with *Bacillus thuringiensis* strain EAPL24 16S ribosomal RNA gene, partial sequence and GCU-*Bt* 25.1/1 showed 99% homology with *Bacillus thuringiensis* strain EAPL05 16S ribosomal RNA gene, partial sequence.

The amplified 276bp conserved region of *cry1* gene of all the 5 *Bt* isolates (GCU-*Bt* 1.5/1, GCU-*Bt* 3.1/1, GCU-*Bt* 6.1/1, GCU-*Bt* 11.1/2, GCU-*Bt* 14.1/1 and GCU-*Bt* 25.1/1) showed homology with *Bacillus thuringiensis* strain. GCU-*Bt* 1.5/1 showed 97% homology with JN135249.1 *Bacillus thuringiensis* strain SSy125-c clone HA1 delta-endotoxin *cry1A* gene, partial cdsLength=3545. GCU-*Bt* 3.1/1 showed 98% homology with FJ617446.1 *Bacillus thuringiensis* strain Tm41-4 *cry1Ac*-like protein *cry1Ac* gene, partial cds Length=3532. GCU-*Bt* 11.1/2 showed 98% homology with GU294785.1 *Bacillus thuringiensis* strain HD07 *cry1Ac* endotoxin gene, complete cds Length=4202.

GCU-*Bt* 14.1/1 showed 97% homology with EU623975.1 *Bacillus thuringiensis* strain LSZ9408 *cry1Ac* gene, complete cds Length=3537. GCU-*Bt* 25.1/1 showed 99% homology with DQ285666.1 *Bacillus thuringiensis* delta-endotoxin *cry1Ac* gene, complete cds Length=3734.

Among six *B.t.* isolates, GCU-*Bt* 25.1/1 was found the most toxic with LC₅₀ 168.8 µg/ml and was isolated from moist soil containing leaf litter. Protein profile of delta-endotoxin protein was performed showing 130 kDa protein band which probably represents *cry1* proteins.

Ecologically *Bt* is safe, Since the *Bt* Toxins are highly specific against certain insects without affecting predators and other beneficial insects. Understanding of the nature of cry gene expression has led to the production of many beneficial pesticides and with knowledge of transformation and genetic manipulation, a foundation has been laid for potentially more effective biopesticides. It has become well recognized that cry-based pesticides generally have low costs for development and registration. The cost of *Bt* pesticides is estimated at 1/40th that of a comparable novel synthetic chemical pesticide.