

B-89: Cloning of insecticidal crystal protein gene from *Bacillus thuringiensis* var. *aizawai* HD 133 and local strain 14j in *Escherichia coli* K-12.

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Six selected *Bacillus thuringiensis* (Bt) strains among 200 strains collected from Sri Lanka, were tested for toxicity for lepidopterans. By strain serotype 7 *aizawai* - HD 133 obtained from University of Cambridge was used as a standard test strain.

Bt strains are of great interest in environmentally friendly pest control programme as they are used in commercial formulations for control of lepidoptera and diptera. ICP gene in Bt is expressed in the stage II of the sporulation and the synthesis continues until the late sporulation. Recombinant strains of *E. coli* is known to express ICP gene at all stages of growth.

We have cloned 3.8-4 kb fragment containing ICP gene in *E. coli*. Total plasmid DNA isolated from var. *aizawai* HD-133 and 14j strain were restricted with Dra I. Purified DNA fragment of 3.6- 4 kb was ligated to pUC 18 restricted with Sma I. Recombinant plasmids were transformed to *E. coli* K-12. The 6.3 kb plasmid with ICP gene in polycloning site inactivated the aminoterminal fragment of β galactosidase and abolished the α -complimentation. Those white colonies were selected as recombinant *E. coli* carrying ICP gene. Expression of ICP was confirmed by phase contrast microscopy and toxicological studies with *Bombyx mori*.