E9

Isolation and Characterization of Novel Insecticidal cry1-Type Genes from Bacillus thuringiensis Strains

Ming Shun Li, Jae Young Choi, JongYul Roh, Hee Jin Shim, Joong Nam Kang and Yeon Ho Je

School of Agricultural Biotechnology, Seoul National University, Seoul, 151-742, Korea.

To identify new *cryl*-type insecticidal genes, 56 lepidopteran-specific Bacillus thuringiensis (Bt) strains were isolated from Korea. Universal oligonucleotide primers, K5un2 and K3un2, were designed and used to amplify all known cryl-type genes with PCR from 56 Bt isolates. The restriction fragment length polymorphism (RFLP) patterns of the PCRamplified fragments revealed 7 distinct patterns, and a standard isolate was selected from each one of the RFLP groups. Through cloning and sequence analysis of the PCR-amplified fragments showing unique RFLP pattern, 7 novel crv1-type genes were identified. Bt K-1 and Bt 2385-1 strains were selected to isolate and characterize the active regions of the novel genes, and then 5 novel genes were cloned from them. To verify the activity against lepidopteran larvae, the novel genes were expressed using baculovirus expression vector system and the bioassay was performed to Plutella xylostella (Px) and Spodoptera exigua (Se). All novel cryl-type genes had high toxicities against Px larvae and, cry1-44 and cry1-5 had the highest toxicity against Px and Se larvae, respectively. Also, cry1-1 and cry1-5 had dual toxicities against Px and Se larvae. Finally, each novel gene must be a very useful resource for development of microbial insecticide and insect resistant plants.