Thesis Title : GENE TRANSFER SYSTEM IN *BACILLUS THURINGIENSIS SUBSP. ISRAELENSIS* BY CONJUGATION-LIKE PROCESS

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ABSTRACT

Protoplast of a lysozyme sensitive strain of *Bacillus sp.* strain O 016 was transformed by plasmid pTF6 and pHV33 with the frequencies of 3.2×10^3 and 2.0×10^3 transformants/µg DNA respectively. The plasmids pTF6 and pHV33 in *Bacillus sp.* strain O 016 was subsequently transferred to *Bacillus thuringiensis* subsp. *israelensis* strain A084 (*B.t.i.*A084) by conjugation-like process with the frequencies of 3.5×10^{-7} and 2.5×10^{-7} (number of transcipients per number of recipients), respectively. Plasmid pTF6 was found to be stably maintained in *Bacillus sp.* strain O 016 and *B.t.i.*A084 but the plasmid pHV33 was found to be unstable in *Bacillus sp.* strain O 016 and B.t.i.A084.

Recombinant plasmids, pTF6-Xd and pHV33-Xd which were constructed by ligation between plasmid pTF6

or pHV33 and the xylosidase gene were transferred into *Bacillus sp.* strain O 016. It was found that plasmids pTF6-Xd and pHV33-Xd were not stable in either hosts.

Another recombinant plasmids, pBA401 which composed of plasmid pTF6 and the pencillin acylase gene was transferred into *Bacillus sp.* strain O 016 and subsequently into *B.t.i.*A084 with the frequencies of 9.9×10^{2} transformants/μg DNA and 6.7×10^{-7} (number of transcipients per number of recipients),respectively. Plasmid pBA401 was found to be stable in both *Bacillus sp.* strain O 016 and *B.t.i.*A084. Studies by using hybridization technique indicated the presence of plasmid pBA401 in *Bacillus sp.* strain O 016. The gene for penicillin G acylase was also found to express in both *Bacillus sp.* strain O 016 and *B.t.i.*A084 hosts.