3-4-17. Expression of a Fusion Protein Between Cry1Ac and Green Fluorescent Protein in an Acrystalliferous Bacillus thuringiensis

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Expression of a fusion gene comprising B. thuringiensis crystal protein, cry1Ac1 and green fluorescent protein (gfp) genes in B. thuringiensis Cry-B strain was examined. The cry1Ac1 gene was cloned in B. thuringiensis-E. coli shuttle vector, pHT3101, under the control of the native cry1Ac1 gene promoter and gfp gene was inserted at the XhoI site which is located after the proteolytic cleavage site, in the middle of the cry1Ac1 gene (pProAc-GFP). B. thuringiensis Cry-B strain carrying pProAc-GFP (ProAc-GFP/CB) produced mRNA transcripts encoding the cry1Ac1-gfp fusion gene but did not produce an inclusion body. Although its expression level was relatively low, this recombinant strain expressed the fusion protein. However, immunoblot analysis, using GFP and Cry1Ac1 antibodies, demonstrated that the fusion protein was not a single species, but multiple ones with various sizes. Besides, the N-terminal fragment of Cry1Ac1 and an independent GFP were also found in B. thuringiensis Cry-B sporulation. Sporulated cells and spore-crystal mixtures ProAc-GFP/CB had insecticidal activity against Plutella xylostella larvae. These results suggest that the fusion protein including a B. thuringiensis crystal protein may be functionally able to be expressed in *B. thuringiensis*.