Construction and Characterization of Transformed Insect Cells Expressing Antibacterial Protein or Baculovirus Very Late Factor-1

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Transformed insect Sf9 cells expressing antibacterial protein (Nuecin) or baculovirus very late factor (VLF-1) were constructed by using *Autographa californica* nuclear polyhedrosis virus (AcNPV) immediate early gene (*ie1*). Neomycin resistant gene as a selection marker was introduced under the control of AcNPV *ie1* promoter, and *nuecin* or *vlf-1* was introduced under the control of the heat shock protein promoter to yield dual expression plasmid with two independent transcription units. It was transfected into Sf9 cells and cell clones expressing Nuecin or VLF-1 were selected by G418 (1 mg/ml) treatment. Genomic DNA from the transformed cells was isolated and integration of AcNPV *ie1* harboring *nuecin* or *vlf-1* was confirmed by PCR and Southern blot analysis. Nuecin was successfully expressed in the transformed cells and secreted into media. Antibacterial activity of Nuecin secreted from the cells was tested against *Escherichia coli*. Thus, this result revealed that the transformed insect cells do not require antibacterial protein in the cell culture system. Furthermore, the transformed cells expressing *vlf-1* in an infection-independent manner were expressed foreign gene products of recombinant baculovirus in the early stage compared with control Sf9 cells. It should also be possible to develop highly efficient transformed insect cells for baculovirus expression vector system.