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# Construction and characterization of transformed insect cells that do not require antibacterial protein in the cell culture system

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Transformed Sf9 cells expressing antibacterial protein 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 antibacterial protein gene (*Nuecin*) 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 were selected by G418 treatment. Genomic DNA from transformed cells was isolated and integration of AcNPV *ie1* harboring *Nuecin* was confirmed by PCR using AcNPV *ie1*-specific primers. The Nuecin was successfully expressed in the transformed cells and secreted into media. Antibacterial activity of Nuecin secreted from the transformed 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.