Construction of Transfer Vectors for Production of Baculovirus Occlusion Bodies that Contain Novel Recombinant Crystal Protein

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Baculovirus occlusion bodies have been recently engineered to incorporate foreign protein such as the *Bacillus thuringiensis* Cry1Ac protein for improvement of insecticidal activity. In this study, *polyhedrin*, *cry1Ac*, *egfp* and *cry1Ca* genes were fused to produce occlusion bodies that contain novel recombinant crystal protein by homologous recombination between *cry1Ac* and *cry1Ca* genes in insect cells. The *cry1Ac*, *cry1Ca* and *egfp* genes were amplified by PCR from pProAc, pBacPHcry1C and pEGFP as the templates, respectively. The amplified genes were fused in pGEM5Zf(-) vector in two orders, *cry1Ac?egfp?cry1Ca* and *cry1Ca?egfp?cry1Ac*. These full fusion genes were transferred into restriction sites, *Xho* I and *Not* I, at the back of *polyhedrin* gene of pOB I (named as transfer vectors, pBacPAC-F and pBacPCA-F, respectively). The fusion constructs were confirmed by restriction endonuclease analysis, DNA sequencing and PCR. The transfer vectors, pBacPAC-F and pBacPCA-F were cotransfected with bApGOZA in Sf9 cells and the recombinant viruses were confirmed by PCR.