Recombinant Baculovirus-Based Multiple Protein Expression Platform for *Drosophila* S2 Cell Culture

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We developed a platform for selective and controllable expression of multiple foreign protein types in insect cell culture. Based on the fact that baculovirus cannot replicate in nonpermissive *Drosophila melanogaster* Schneider line 2 (S2) cells, S2 cells that stably express hEPO under the control of the S2-derived inducible metallothionein promoter were infected with three types of recombinant baculoviruses, each of which expressed a fluorescent protein (EGFP, ECFP, or EYFP) gene under the control of MT promoter. Addition of $CuSO_4$ as an inducer to infected, stably transfected S2 cells resulted in simultaneous expression of hEPO and three fluorescent proteins. Importantly, expression profiles and levels of hEPO were similar in both non-infected and infected cells, indicating that baculovirus infection did not affect the expression of stably introduced foreign genes. Expression of the three fluorescent proteins was able to be selectively regulated by altering combination ratios of the three types of recombinant baculoviruses. Such a system would be expected to be attractive as a multiple protein expression platform for glycosylation pathways.