

## Evaluation of viral genes to stabilize transient transgene expression

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### Abstract

Gene expression is a highly regulated and complex phenomenon in eukaryotes. Over-expression of a gene does not always result in an increase in protein levels, but may cause transcriptional or post-transcriptional gene silencing, where the introduced gene and/or its native homologue are suppressed. This can occur with high expressing transgenes and with some virus genes during viral infection. In order to study the process, we used a transient expression system and the *green fluorescent protein (gfp)* gene driven by the constitutive 35S promoter. Following introduction of the *gfp* reporter gene, GFP expression reaches a maximum intensity at 24 hours post-introduction and declines rapidly thereafter. For this research, we evaluated 6 different virus genes known to suppress gene silencing, in an attempt to stabilize GFP expression over time. Among the six silencing suppressors evaluated, HC-Pro and p19 had a stabilizing affect GFP expression over 100 hours post-introduction when introduced as 3' fusions with GFP (*cis*). In contrast,  $\gamma$ b and p21 stabilized GFP expression for over 100 hours when they were introduced with GFP on separate expression vectors (*trans*). The remaining suppressors, AL2 and the 126 kDa replicase, did not stabilize GFP expression in our transient system. These data suggest that some silencing suppressors interfere with a portion of the silencing pathway involved in transgene silencing. This research highlighted the potential utility of viral suppressors of silencing, when used in the correct combination with the gene of interest, to improve plant transformation efficiencies and stabilizing transgene expression.