Abstract

We constructed HIV mutants by replacing the matrix domain of Gag with the protease (PR) domain. The resulting construct was cloned into an HIV replication-defective vector, which carries a gpt gene in env region and an inactivated PR in the pol gene. The chimeric construct MA(PR) was unable to produce particles when expressed alone in 293T cells. This mutant could also transdominantly interferring with wild-type virus particle production when co-expressed with a wild-type (wt) Gag protein expression plasmid. The trans interference effect of the mutant on wt virus budding was dose-dependent. Virus particle production was abolished when cotransfected DNA of Gag versus MA(PR) was at a ratio of 1:1. However, levels of released virus particles were increased when the MA(PR) DNA amount used for cotransfection was reduced. MA(PR) could be efficiently incorporated into wt Gag particles based on in vitro reverse transcription (RT) assay when cotransfected plasmid DNA ratio of Gag/MA(PR) was kept at 20:1 to 10:1. Although the MA(PR) was incorporated into virus particles at a level comparable to that of wt, the PR within the context of MA(PR) did not appear to be functional appropriately since the Pr55gag represented the major form of released virus particles. This suggests that sequences outside the PR domain may be involved in the process of PR activity regulation and that the a proper position may also affect the acquisition of PR enzymatic activity.

Key Word：Chimeric virus-like particles、protease、Gag