Development of a T7 RNA polymerase expressing cell line using lentivirus vectors for the recovery of recombinant Newcastle disease virus

ABSTRACT

The development of a T7 RNA polymerase (T7 RNAP) expressing cell line i.e. BSR T7/5 cells marks an improvement of reverse genetics for the recovery of recombinant Newcastle disease virus (rNDV). BSR T7/5 is developed by transient transfection of plasmid encoding T7 RNAP gene for rNDV rescue. However, the gene expression decreases gradually over multiple passages and eventually hinders the rescue of rNDV. To address this issue, lentiviral vector was used to develop T7 RNAP-expressing HEK293-TA (HEK293-TA-Lv-T7) and SW620 (SW620-Lv-T7) cell lines, evidenced by the expression of T7 RNAP after subsequent 20 passages. rNDV was rescued successfully using HEK293-TA-Lv-T7 clones (R1D3, R1D8, R5B9) and SW620-Lv-T7 clones (R1C11, R3C5) by reverse transfection, yielding comparable virus rescue efficiency and virus titres to that of BSR T7/5. This study provides new tools for rNDV rescue and insights into cell line development and virology by reverse genetics.

Keyword: Reverse genetics; Lentiviral vectors; Cell line development; T7 RNA polymerase-expressing cell lines; Recombinant Newcastle disease virus