

P019 Membrane active peptide enhances Tat-mediated gene delivery

Saleh A. F. , Aojula H, Pluen A

*School of Pharmacy and Pharmaceutical Sciences,
University of Manchester, UK*

In an attempt to enhance the transfection efficiency of Tat-derived peptide as a gene carrier, a membrane active peptide (LK15) was covalently attached to Tat to construct the composite peptide (LK15-Tat). Plasmid DNA (pDNA) condensation was assessed by YOPRO-1 fluorescence quenching assay. Gene transfer efficiency of LK15-Tat was compared to Tat and LK15 in two cancer cell lines (H1080 and HT29) using a luciferase reporter gene. Cellular uptake of peptide/pDNA-FITC complexes was investigated by flow cytometry. Efficient pDNA condensation by LK15-Tat was observed at a (+/-) charge ratio of 2:1 and above. LK15-Tat displayed remarkably higher level of gene transfer compared to Tat or LK15 in both cell lines examined. In HT1080 cells, gene expression mediated by LK15-Tat was not affected by the presence of chloroquine, while a 2-fold increase was observed in HT29 cells. LK15-Tat/pDNA complexes exhibited higher cellular uptake than Tat- or LK15/pDNA complexes in both cell lines. These results suggest that attachment of LK15 to Tat may enhance the transfection efficiency in part due to improving the endosomal escape and/or the cellular uptake of pDNA complexes. LK15-Tat represents a potential agent for effective peptide-based gene transfer into mammalian cells.