Designing and constructing a vector-based microRNA to knockdown STAT3 gene to target breast cancer stem cells

Sara Noorani[*1], Mehdi Forouzandeh Moghaddam[1], Fatemeh Salimi [1]

1. Medical biotechnology department, school of medical sciences, Tarbiat Modares University, Tehran, Iran

sara.noorani@modares.ac.ir

microRNA is an attractive tool for future therapies which regulates gene expression by binding to target mRNA and depending on the degree of complementary binding, regulates gene expression by either repression of translation or degradation of mRNA. STAT3 transcription factor has been activated inappropriately in variety of cancers and regulates genes involved in proliferation, survival, self-renewal, invasion, and angiogenesis. Considering the significant roles of STAT3 in breast cancer stem cell, it is expected that silencing STAT3 could have a dramatic effect on decreasing this population and inducing apoptosis in tumor cells. Since endogenous microRNAs are powerful tools to silence gene at post-transcriptional level we intended to design and clone an artificial microRNA specific to STAT3 gene. The designed miRNA mimics flanking and loop sequences of endogenous miR-155 structures but stem-duplex is specific for target sequence. Upon expression inside the cell, this miRNA can be spliced by natural miRNA splicing enzymes Drosha and Dicer to form a mature miRNA. This mature miRNA potentially silence STAT3 gene. The microRNA was ligated to the miRNA expression vector pcDNA6.2 which controls miRNA expression by CMV promotor. Ligated vector was transformed into E.coli TOP10 and colony selection was performed to insure the efficiency of transformation.

Key words: STAT3, miRNA, breast cancer stem cell