Functionalizing liposomes with anti-CD44 aptamer for selective targeting of cancer cells

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ABSTRACT

CD44 is a glycoprotein receptor known to play a role in cell adhesion, growth, and motility. CD44 is found to be overexpressed by many tumors and is identified as one of the most common cancer stem cell surface markers including tumors affecting colon, breast, pancreas, and head and neck, making this an attractive receptor for therapeutic targeting. In this study, 2'-F-pyrimidine-containing RNA aptamer (Apt1), previously selected against CD44, was successfully functionalized to the surface of PEGylated liposomes (Lip) using the thiol-maleimide click reaction. The binding affinity of Apt1 was improved after conjugation compared to free-Apt1. The cellular uptake for Apt1-Lip was tested by flow cytometry and confocal imaging using the two $CD44^+$ cell lines, human lung cancer cells (A549) and human breast cancer cells (MDA-MB-231), and the $CD44^-$ cell line, mouse embryonic fibroblast cells (NIH/3T3). Furthermore, siRNA targeting luciferase gene (Luc2) has been trapped inside the preformed liposomes (Lip-siRNA) and then the Lip-siRNA functionalized with Apt1 using post-insertion method. The efficacy of Apt1-Lip-siRNA nanoparticles in knocking down of Luc2 expression were tested in MDA-MB-231-Luc2-GFP breast cancer cells in vitro and showed higher inhibition to Luc2 expression comparing to Lip-siRNA alone. Furthermore, orthotopic xenograft model of human breast cancer was developed by implanting the MDA-MB-231-Luc2-GFP cells into female nude mice and our siRNA delivery system successfully knowckdown the Luc2 expression in vivo. In conclusion, we demonstrate a successful conjugation of anti-CD44 aptamer to the surface of liposome and the binding preference of Apt1-Lip to the CD44-expressing cancer cells which conclude a promising potency of Apt1-Lip as a selective drug delivery system.