Comparison of the efficiency of PEG and SPDP crosslinkers used for mAb conjugated 5-fluorouracil loaded BSA nanoparticles

G. Fadaeian, S.A. Shojaosadati, H. Kouchakzadeh

1 Biotechnology Group, Faculty of Chemical Engineering, Tarbiat Modares University, Tehran, Iran. shoja_sa@modares.ac.ir

Abstract

Site-specific delivery of drugs to tumor cells can significantly reduce drug toxicity and increase their therapeutic effect due to higher drug levels in tumor tissue. Among different drug delivery systems, protein nanoparticles have gained much interest due to their inherent properties of biodegradability, lack of toxicity and nonantigenicity[1]. In this study, Bovine Serum Albumin (BSA) nanoparticles containing anticancer drug 5-Fluorouracil (5-FU) were synthesized. Polyethylene glycol (PEG) conjugation of nanoparticles (PEGylation) was performed according to our previous study [2]. Then, anti-HER2 monoclonal antibody, 1F2, was conjugated to 5-FU loaded BSA nanoparticles through two well-known heterobifunctional crosslinkers MAL-PEG5000-NHS and SPDP (N-Succinimidyl 3-(2-pyridyldithio)-propionate). Efficiency of 1F2 coupling on the surface of BSA nanoparticles by using different crosslinkers was investigated by Enzyme-linked immunosorbant assay (ELISA) and flow cytometry methods.

mAb conjugated to BSA nanoparticles with PEG and SPDP was characterized by dynamic light scattering (DLS) and show average particle size of 150 and 130 nm, zeta potential of 22.4 and 25 mv and PDI of less than 0.06 respectively.

In vitro cumulative release of 5-FU from BSA nanoparticles was investigated in PBS solution, pH 7.4 at 37 °C. An initial burst release of drug from non-PEGylated and PEGylated BSA nanoparticles was observed which is related to the adsorbed 5-FU on the surface of nanoparticles. In the case of drug loaded nanoparticles, 5-FU was released totally during 48 hours but in PEGylated BSA NPs the maximum release within 48h was approximately 80%. This results reveal slower release of 5-FU from PEGylated BSA nanoparticles as compared with non-PEGylated BSA NPs(figure.1)

ELISA results show that approximately 20% of Anti-HER2 mAb 1F2 were conjugated to nanoparticles which are equivalent to approximately 100% of NH2 group by using hetrobifunctional PEG. The results obtained for SPDP linkers are much lower than results of PEG. This 5-FU targeted drug delivery system is under in vitro and in vivo studies. In addition, stability studies of produced 5-FU delivery system proved its high immunoreactivity during three months. This novel HER-2 targeted 5-FU delivery system can be employed for improvement in breast cancer treatment.

References

Figure 1. *In vitro* cumulative release of 5-FU from albumin nanoparticles in PBS solution, pH 7.4 at 37 °C.