SYNTHESIS OF RITUXIMAB IMMUNOCONJUGATES FOR SUBSEQUENT RADIOLABELING

Darinka Gjorgieva Ackova, Katarina Smilkov, Emilija Janevik - Ivanovska

[1] Faculty of Medical Sciences, Goce Delčev University, Štip, R. Macedonia
e-mail: darinka.gorgieva@ugd.edu.mk

Introduction

In order to increase the therapeutic efficacy unmodified monoclonal antibodies (mAbs) are associated with drugs, toxins and radionuclides. This immunoconjugates are examples of mAbs specifically designed to deliver their toxic loads directly to cancer cells. In general, one target-specific radioimmunoconjugate consists of three parts: biomolecule-carrier (mAb), bifunctional chelating agent (BFCA) and radionuclide (radiometal, M) (Fig. 1). The development of pharmaceutical formulation of an antibody "ready to label" with different radioisotopes (Lu-177, Y-90), we believe that can be of a major clinical impact.

The aim of this work was to perform conjugation of mAb rituximab, with three different BFCAs, *p*-SCN-Bn-DOTA, *p*-SCN-Bn-DTPA and 1B4M-DTPA in a form of ready-to-label kit formulations.

Experimental

Conjugation of rituximab with selected BFCA's, was made by mixing the purified antibody diluted in phosphate buffer saline (0.1 M, pH = 8.0) and appropriate BFCA in ratio 1:20. Mixture was incubated 18 hours at 2-8 °C, with constant slight stirring. The resulting rituximab immunoconjugates were purified with ultrafiltration and subjected to the lyophilization process by using Labconco Free Zone Stoppering Tray Dryer, (USA) (Fig 2).

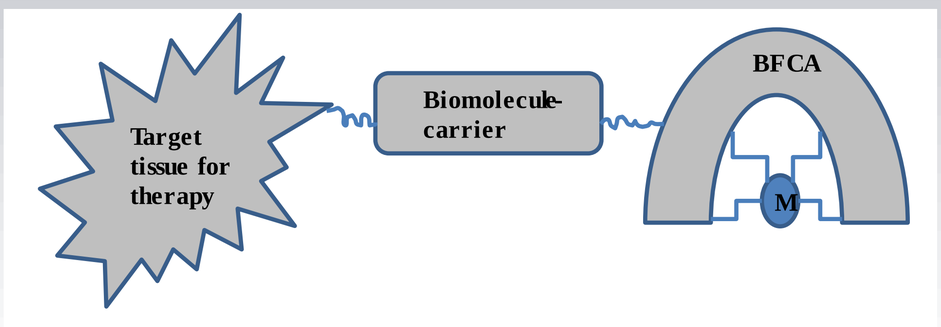

Fig.1. Schematic representation of radiopharmaceutical components

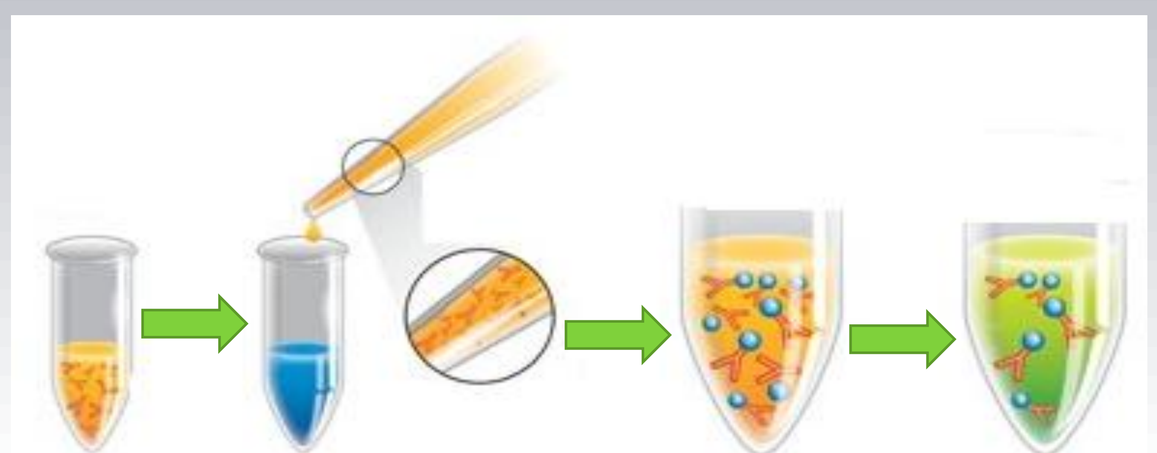

Fig. 2. Protocol for preparation of lyophilized formulations

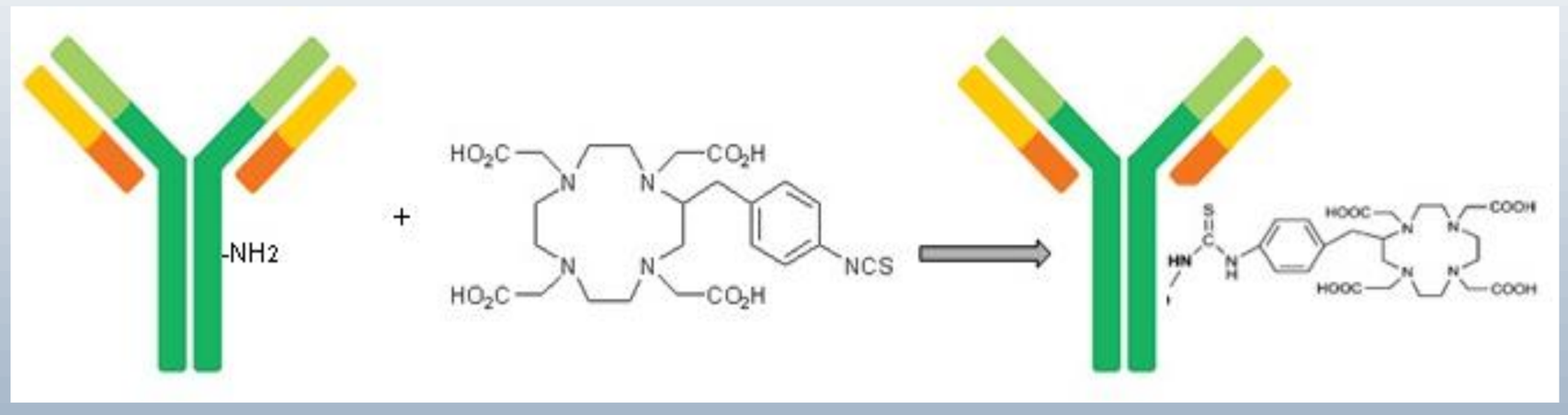

Fig. 3. Schematic representation of the conjugation reaction of rituximab with *p*-SCN-Bn-DOTA.

Fig. 4. Immunoconjugates of different BFCA's in lyophilized state.

Results

The BFCA's used for conjugation are derivatives of tetraazacyclododecane tetraacetic acid (DOTA) and diethylenetriamine pentaacetic acid (DTPA). The conjugation process undergoes reaction of the active ester group of the chelator with the neutral form of the amines of the antibody. Specifically, bonds of thiourea type with ε-amino groups of lysine residues of the antibody are formed. The resulting rituximab immunoconjugates were purified with ultrafiltration and subjected to the lyophilization process by using Labconco Free Zone Stoppering Tray Dryer, (USA).

Conclusions

These successfully synthesized immunoconjugates can be a good basis for conducting further experiments with radiolabelled formulations in order to develop promising new radiopharmaceutical.

***XXIV Congress of Chemists and Technologists of Macedonia, 11-14 Sep 2016, Ohrid, R. Macedonia**