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The conjugates of HER2 and integrin $\alpha_v\beta_3$ binding proteinsfor use in anti-cancer therapies.

Abstract

One of the most commonly used types of cancer treatment is chemotherapy. Because of the specificity for dividing cells and not only cancer cells, chemotherapy is responsible for many significant adverse effects influencing the quality of life of patients and the overall effectiveness of treatment. An attempt to combine high efficacy of the treatment with its specificity affecting the limitation of side effects is the use of monoclonal antibodies combined with strong cytostatics in the form of antibody-drug conjugates called ADCs.

In the present study we focused on developing an alternative for the ADCs, wherein the antibody has been replaced by a protein having similar properties, but mainly differing in size and structure complexity. The purpose was to limit the defects of ADCs resulting from the presence of antibodies - mainly on reducing costs and facilitating production.

In this study we engineered two conjugates - diaffibody anti-HER2 and scFv anti-integrin $a_v\beta_3$ - with the drug monomethyl auristatin E (MMAE), synthetic strong cytostatic which is a derivative of the naturally occurring peptide in the marine shell-less mollusc *Dolabella auricularia*, inhibiting cell division by blocking tubulin polymerization.

As a part of the work we designed, overexpressed and purified both proteins, diaffibody anti-HER2 and scFv anti-integrin $a_v\beta_3$, and performed their biophysical characteristics intending mainly to explore their interactions with the ligands. In the next stage the protein were subjected to conjugation to a cytostatic drug, and the conjugates were subjected to further analysis.

The potential antitumor activity of the conjugates was analyzed. We performed the analysis of their effect on the viability of tumor cells characterized by the overproduction of ligand, relative to control cells. The diaffibody anti-HER2-MMAE conjugate demonstrated relatively high cytotoxicity, causing a decrease in HER2-positive cell viability up to 10%. The calculated IC_{50}

parameter was 1 – 1 nM depending on cell line. The weaker results were obtained for the scFv anti-integrin $a_v\beta_3$-MMAE conjugate, causing integrin $a_v\beta_3$-positive cells viability decrease up to 20%, but requiring the use of relatively high concentrations. The calculated IC_{50} parameter was 115 nM.

The conjugates obtained in this work have the potential for use in anti-cancer therapies.

Keywords:

anti-cancer therapies, HER2, integrin $\alpha_v\beta_3$, scFv, affibody, conjugates