Superparamagnetic nanoparticles - synthesis, characterization and functionalization using biological macromolecules

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Abstract

Recently, nanotechnology has spread into many areas of science including medicine, pharmacology and biotechnology. The diversity of nanoparticles enabled development of innovative technologies in drug formulation, targeted therapy, drug delivery and diagnostics. Nanomaterials such as nanoparticles display unique physical, optical and chemical properties that in combination with large surface to the volume ratio make them useful for cancer diagnosis and treatment.

Superparamagnetic nanoparticles belong to the group of nanomaterials that are intensively explored in oncology. After applying the magnetic field they produce the heat thanks to Neel and Brown relaxation. These phenomena, combined with targeting agents and cytotoxic drugs, have potential to improve cancer treatment.

In this study the multifunctional superparamagnetic nanoparticles were constructed. In the first step the synthesis of dextran-coated iron oxide nanoparticles (IONP@Dex) was optimized followed by conjugation with targeting agent - anti-Her2 aptamer. The specificity of the targeted nanoparticles was determined by studying their biological effect relative to that of non-tagged nanoparticles against two cell lines, human adenocarcinoma SK-BR3, overexpressing the Her2 receptor, and U 87-MG, human glioblastoma epithelial

cell line, not overexpressing Her2. In order to confirm the interaction of the nanoparticles with cells, we used fluorescence microscopy and FACS analysis. The experiments showed that the aptamer-conjugated nanoparticles were highly specific towards the Her2-expressing cells. In the next step the nanoparticles were used in targeted hyperthermia. SK-BR3 and U-87 MG cell lines were treated with non-conjugated and aptamer anti-Her2 conjugated nanoparticles followed by applying the magnetic field. After that the cytotoxicity test was performed. The results revealed that hyperthermia performed by using the nanoparticles conjugated with aptamer is ca. 90-fold more effective than by using non-conjugated nanoparticles.

In the second part of the research nanoparticles were conjugated by hydrolysable linkers with cytotoxic drugs, doxorubicin and momomethylauristatin E. The results of the conjugation reaction were evaluated by fluorescence spectrum measurement for doxorubicin, since it reveals fluorescence properties; and by mass spectrometry (in case of MMAE). The cytotoxicity tests showed that conjugates of nanoparticles and cytotoxics are at least two times more toxic than free drugs. The nanoparticles–MMAE conjugates were used in hyperthermia experiment, but no significant effects were observed.

The last part of the work dealt with constructing multifunctional nanoparticles, containing the cytotoxic drug and targeting agent selective against cancer cells. As a targeting agent, the anti-Her2 affibody was chosen. The affibody belongs to the group known as “alternative scaffolds” and is characterized by high stability and strong affinity to the receptor. Both the affibody and monomethylauristatin E were conjugated to dextran-coated nanoparticles. The selectivity and effectiveness of the conjugates were evaluated using SK-BR3 and U-87 MG cell lines. The viability tests revealed that the nanoparticles were specific to Her2-

overproducing cells, which means that the MMAE as well as the affibody were conjugated to the nanoparticles in a proper way.

In summary, the presented results clearly show the anticancer potential of superparamagnetic nanoparticles. The methods of conjugation with targeting agents and cytotoxic drugs were developed and optimized. These findings can improve new anti-cancer strategies which are based on nanotechnology.