Drug Delivery Using Biodegradable Tyrosine-based Nanospheres

Larisa Sheihet, Robert A. Dubin, David Devore and Joachim Kohn
Department of Chemistry and Biological Chemistry, Rutgers, the State University of New Jersey

Statement of Purpose: It is widely recognized that nanotechnology will have a profound effect on the delivery of pharmaceuticals with poor bioavailability by improving their stability, circulation times in the body, and permeability through cell membranes, while reducing their toxicity.[1] A majority of all drugs, including anti-tumor agents, anti-depressants, and statins, are hydrophobic and therefore require a solubilization process to enable their parenteral delivery.[2] Of the many alternative approaches proposed to overcome this obstacle, perhaps the most promising is the use of amphiphilic block copolymers that self-assemble into supramolecular nanoparticle delivery vectors.[3] Our approach in this area has been to develop a biodegradable polymer with a high degree of structural versatility enabling self-assembly into nanospheres that can deliver a wide array of hydrophobic compounds.[4,5] These nanospheres are designed to avoid adsorption of proteins and adhesion to cells, and to limit recognition and elimination by the reticuloendothelial system.[6] Here we describe the synthesis and evaluation of an extended family of amphiphilic ABA-triblock copolymers that consist of hydrophilic A-blocks of poly(ethylene glycol) and hydrophobic B-blocks of desaminotryosyl-tyrosine esters (DTR) and diacids. We demonstrate that these form stable nanospheres that complex a wide variety of hydrophobic therapeutic agents and deliver them efficiently to cells in vitro.

Methods: The triblock copolymers were synthesized as previously described[5] and self-assembly of the copolymers into nanospheres was induced using a static method method. Physico-chemical properties of the triblock copolymers, the resultant nanospheres and drug association efficiency were assessed by ^{1}H NMR, DSC, GPC, HPLC and DLS techniques. The in vitro cellular activities of several representative anti-tumor drug-nanosphere complexes were determined in the cervical carcinoma cell line KB using an MTS assay.[5] Nanosphere toxicity was determined in vivo using non-tumor-bearing immune competent mice to obtain the maximum-tolerated dose (MTD).[7]

Results / Discussion: The degree of hydrophobicity of the middle blocks (B) was systematically varied depending upon the length of the diacid and/or the nature of the pendent group R of the DTR monomer, while the PEG end blocks were kept constant (Figure 1).

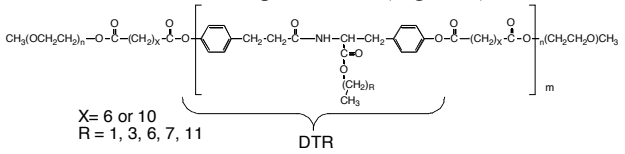

Figure 1. Tyrosine-derived triblock copolymers.

The hydrodynamic diameters of the resulting nanospheres were dependent on the copolymer hydrophobicity and the method of producing the nanospheres.

The nanospheres formed stable complexes with chemotherapeutic agents. Complexation efficiency was found to be dependent on the oil:water partition coefficients of the drugs, log D (Figure 2). The in vitro KB tumor cell LC50 values for the nanosphere-drug complexes were essentially equivalent to those of the free drugs.

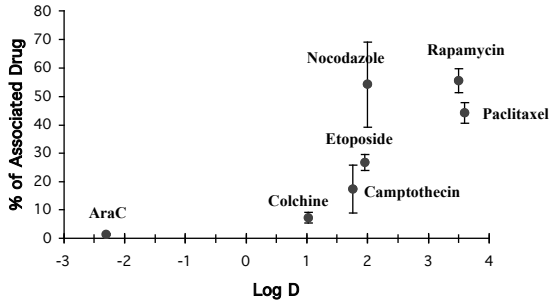

Figure 2. Nanosphere complexation of drugs depends on the drug partition coefficients, log D

Conclusions: An evaluation of the tyrosine-based amphiphilic ABA triblock copolymers nanospheres exhibited preferential strong complexation with hydrophobic materials. Our results suggest that the nanospheres act as an effective sink for absorbing and binding hydrophobic drugs and the association efficiency increases with drug hydrophobicity and/or hydrophobicity of the middle block of the nanospheres. The delivery of chemotherapeutic agents with varying Log D values to the tumor cells in vitro was confirmed by growth inhibition of KB human carcinoma cells.

References:

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