Lipid Carriers for Genetic Drugs in Gene Therapy

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Abstract: The chemical nature of the genetic drugs (*e.g.* antisense oligonucleotides, siRNA, vectors) requires suitable carrier system that would protect them from enzymatic degradation without changing their properties and enable efficient delivery into targeted cells. Lipid vectors for nucleic acid delivery that have been widely investigated for years can be very effective in many cases. Enormous diversity of natural and synthetic lipids as well as polymers allow the manipulation of carrier parameters (particle size, surface charge, cell or tissue specificity, stability in defined conditions such as pH, temperature *etc.*) in order to minimize of side effects (*e.g.* haemolytic activity, cytotoxicity to nontarget cells) and improvement of drug delivery into sites of destination. The most of trials that are made in cancer gene therapy field are focused on solid tumours. The blood cancer cells have become the aim of our investigations. We used two leukaemia cell lines (HL-60 and Jurkat T) as model systems. It is well known that these types of cells are very difficult for effective transfection. The aim of our study was to construct lipid vector that would enable efficient transfer of genetic drugs into these cells. The obtained results are promising: high levels of transfection of both Jurkat T and HL-60 lines was achieved. The proposed carrier is based on modified preparation procedure of **CCL** (**C**ationic **C**oated **L**iposomes) [1, 2] but lipid composition is original and they are surface-modified and/or surface-modified and antibody-carrying versions. CCL's consist of a core arising from complexation of positively charged lipid (DOTAP, (1,2-dioleoyl-3-trimethylammonium-propane) and negatively charged plasmid DNA or antisense oligodeoxynucleotides coated by the lipid bilayer composed of PC (phosphatidylcholine), DSPE-PEG (1,2-distearoyl-*sn*-glycero-3-phosphoethanolamine-N-[amino(polyethylene glycol)2000](ammonium salt), DOPE (1,2-dioleoyl-*sn*-glycero-3-phosphoethanolamine) and DC-CHOL(3β-N-[dimethylaminoethane]carbamoyl)cholesterol. The diameter of obtained liposomes was 105 +/- 15 nm and their zeta-potential was -5,4 +/- 1,2mV. The physical properties of the preparation were presented during a last year spring COST Meeting in Lisbon [3].
Here we present the properties of the biological activity i.e. silencing an expression of Bcl-2 gene (protein) expression in leukaemia cells. In this model (cell culture) the construct revealed high effectiveness as was tested on the level of mRNA and protein expression. This carrier was also found to be highly efficient in delivery of plasmid-coding for siRNA's in several cell-line systems.
We also constructed the modification of the above described carrier which was enriched in the synthesised cyanur-PEG-PE serving as an anchor for covalent attachment of the antibodies specific for the leukaemia cell-line markers. Obtained in this way targeted liposomes containing antisense (Bcl-2) ODN's were found to specifically quench Bcl-2 expression in the target cell lines.
Key words: acute leukaemia, cationic lipids, gene therapy, lipid carrier

References:

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