ABSTRACT

The main aim of following work is presenting preliminary studies of the effectiveness of liposomal or lipid formulations preparation, in relation to selected cell models. The task of selected cells is the best possible representation of pharmacokinetic elements including the distribution of substances or carriers in the body and possible future use. The main assumption on which prepared a set of studies is the importance of the process of endocytosis. It is assumed to be a critical step in the pharmacokinetics of nanocarriers. It is known, that after oral or intravenous administration, clearance is the most important factor affecting on amount of active substance available at the target site. Additionally, it is directly related to the rate of drug leakage and the uptake of the nanocarrier itself by the mononuclear phagocyte system (MPS). After reaching to the target cells or to their surroundings, the carrier is collected via endocytotic pathways. The possibility of selecting the endocytotic pathway (involved in the internalization of the carrier) allows targeting to the specific cellular mechanisms, as well as optimization of the carrier in terms of active substance intracellular distribution. In vitro pharmacokinetic evaluation of nanopharmaceuticals requires the selection of appropriate cell lines that reflect the critical elements of biodistribution, elimination or effectiveness in affecting on target cells. The latter element is assessed on the basis of cytotoxicity analyzes. It allows to determine the effectiveness of the active substance, as well as local parameters that must be taken into account at the design stage of a nanopharmaceutical.

It is well known, that the use of nanocarriers allows to reduce the negative side effects of the therapeutics, which is particularly important in the case of cancer diseases. Additionally, it is extremely important to maintain effectiveness of the active substance. In the case of antibiotic resistance, it is necessary to use agents showing no specific molecular targets, against which microorganisms can obtain resistance through genetic mutations. Treatment of a number of comorbidities, including the very popular anemia, requires solving the problem of painful injections of intramuscular iron preparations by improving the absorption of this element from the digestive system. Each of these cases must be considered in the context of the best pharmacokinetic, pharmacodynamic and pharmacogenetic parameters, which should form the basis for research work both at the *in vitro* and *in vivo* level.

The first part of the thesis presents an innovative approach to solving problem of the presence of irritants in popular antiseptics containing octenidine hydrochloride. The lipid-based formulation was tested for antimicrobial activity against *Escherichia coli*. Then, a series of physicochemical analyzes, supported by molecular dynamics, were performed in order to determine the mechanism of action of the active substance, whose reference was the

well-known chlorhexidine. Liposome formulations (neutral, with a composition of 100 mol% POPC and negatively charged, imitating a bacterial membrane, with the composition of 30 mol% DOPG and 70 mol% POPC) have been prepared and used as a model of bilayer.

The second part of the thesis presents results obtained with doxorubicin formulation, which is a widely used anthracycline antibiotic whose anti-tumor activity as well as a number of side effects are well understood. For this purpose, a formulation with the internal structure stiffed by heparin, was used. This polymer was used as a medium to crystallize doxorubicin. This nano-system was tested in context of delivery efficiency, against selected endocytotic pathways. Research based on confocal microscopic examination and molecular biology studies, including real-time PCR and Western Blot method. The aim of these studies was to determine the endocytotic pathways involved in the nanoaggregate uptake. In the next step, cytotoxicity analyzes of the prepared preparations were performer, against the commercial drug Caelyx®. Mapping target cell properties after intravenous supply for doxorubicin (macrophages, breast cancer and liver cancer) was allowed by the respective models.

In the third part of the thesis, a liposomal preparations based on iron salts (inorganic, organic and complex) was prepared. Formulations were analyzed for the encapsulation efficiency, cytotoxicity to the colon cancer cell line (a standard model used in oral administration research) and the capacity of reactive oxygen species generation, which is the main side effect of iron supplementation.

All cytotoxicity and antimicrobial activity tests were carried out in accordance with the relevant normative assumptions, including the Quality Book of Laboratory of Research and Development Center, Regional Specialized Hospital in Wrocław.

The obtained results prove the wide potential of liposomal and lipid nanocarriers to modify the pharmacokinetics and pharmacodynamics of active substances characterized in the work. Appropriate modification of the formulation allows for a targeted therapy, based on the nanopharmaceutical physicochemical properties selection and offers a wide range of possibilities for their use.

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