Tytuł: **Mechanism of incorporation of lipid liquid crystalline drug carriers – cubosomes and hexosomes into lipid membranes**

Mechanizm wnikania ciekłokrystalicznych lipidowych nośników leków – kubosomów i heksozomów w błony lipidowe

Liquid crystalline lipid nanoparticles (LCNP) are considered to have great potential as drug delivery systems and are intensely studied as carriers of especially toxic or ill-soluble drugs. They find interest due to their biocompatibility, ease of preparation and drug incorporation and possibilities of attachment of groups addressing the vehicle to the point of delivery in the organism. The carrier is designed to protect the healthy cells for the toxic side effects of the drug releasing it in the cancer affected cells. Recently we have demonstrated that monoolein cubic phases in the form of gels and dispersed into nanoparticles can hold large amounts of anthracyclines and their release from the carrier can be controlled by pH. The properties of the carrier is highly dependent on both the lipid used for its construction and the stabilizing polymer, e.g. it has been recently reported that phytanthriol based cubosomes are much more toxic than glyceryl monooleate (monoolein) ones and disrupt the membranes of healthy cells without any drug inside. Confocal microscopy images reveal such disruption of membranes thus it was postulated that oxidation stress and changes in membrane organization in case of these carriers are the reason of cell death. In our view, better understanding of molecular mechanism of such interactions is important for the LCNP therapeutic and diagnostic applications and is the goal of our project. Our long-term experience in physical chemistry studies of lipid membranes and interactions with them should be helpful in the realization of the tasks planned in the project. The results of these investigations should allow to design optimal lipid liquid crystalline carriers and their utility in case of anthracycline drugs and methotrexate will be further verified on glioma cancer cells in the cooperating biological and clinical groups of Prof. dr. hab. Marcin Kruszewski in the Institute of Nuclear Chemistry and Technology, and Prof. dr. hab. Leszek Królicki in the Nuclear Medicine Center of Warsaw Medical University.

So far the studies of LCNP drug carriers interactions with lipidic membranes were carried out using supported lipid layer or liposomes as models of cell. The properties of the supported lipid layers depend however on the character and topography of solid state support. In our approach the lipid monolayers spread at the water-air interface (monolayer prepared by the Langmuir-Blodgett technique) will be also used. The flexibility of such membrane – very simplified model of one leaflet of the biological membrane - is not limited by any underlying support. The molecular level description of the interactions of the lipid layer at the air-water interface and following its transfer onto solid support will enable us to determine how the efficiency of adsorption, fusion of the carrier and drug release depends on the properties and organization of the lipid molecules in the membrane, and even more importantly – the optimal composition of the drug carrier for sustainable drug release will be established.

It is known that cancer cell walls are more leaky and permeable than the healthy ones. This property can be resembled by preparing the Langmuir-Blodgett layers at different surface pressures, thus making them more or less condensed. Monitoring changes of surface-pressure and area per molecule with time will allow to evaluate the kinetics of adsorption and fusion processes. Structures and composition of the membranes before and after contact with the drug carrier will be investigated by microscopies (e.g. Brewster angle, and AFM) and spectroscopies (e.g. PM IRRAS) all available in our Faculty and by neutron reflectivity measurements in Grenoble. The processes studied define the stability of LCNP in contact with the lipid membrane and efficiency of drug removal, and therefore the results of these studies will allow to propose to select an optimal system for further biological and clinical studies of sustained drug delivery.