SCANNING ELECTROCHEMICAL MICROSCOPY APPLICATION FOR HYBRID PHOSPHOLIPID BILAYER INVESTIGATION AND MODIFICATION

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Essential part of every living cell is its plasma membrane. Not only it surrounds the cell, but also controls materials transportation and is involved in transmitting a signal coming from the environment [1]. Due to the complex structure of biological membranes, various cell membrane-modeling derivatives have been developed. Solid supported bilayer phospholipid membranes are used as stable phospholipid membranes models for investigating various biologically relevant processes since it exhibits most of the bilayer properties and are quite easy to produce [2]. Widely investigated membrane property is selective permeability, which can be modified. One of the ways to achieve this is electroporation - a physical method in which cells are subjected to an electrical impulse. This results in the formation of pores in the phospholipid layer [3]. Although electroporation is already used in practice (electrochemotherapy, genetic engineering, food industry), certain studies on the size of the pores formed and the time of pore closure are still relevant [4]. By being able to control these process parameters, the phenomenon of electroporation could be applied more extensively and efficiently.

In this work a glass plate coated with a layer of tin oxide doped with fluorine atoms (FTO) was used as a substrate for self-assembled monolayer (SAM) and phospholipid bilayer membrane (BLM) formation. Octadecyl trichloro silane (OTS) was used to form SAM. The hybrid phospholipid bilayer membrane was prepared using the vesicle fusion method from 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC) and cholesterol in phosphate buffer solution (PBS).

In the present research scanning electrochemical microscope (SECM) was applied to investigate hybrid phospholipid bilayer (BLM) surface properties and modify it with electric impulse. Measurements with SECM are widely used in researching or modifying biological systems since it is carried out in a liquid medium that can be easily biocompatible. The SECM signal is a Faradaic current flowing through an ultra-microelectrode (UME) which radius can range from several nm to 25 µm. UME can be moved in space in x, y and z directions which allows to scan the surface and depict it [5]. Through targeted feedback mode approach experiments, SECM was applied to locally detect membrane surface, electroporate it and capture the changes. Reversible and irreversible electroporation was achieved by varying different aspects of method: UME potential vs Ag/AgCl/KClsat, electric impulse time and electrode height above the surface. Since the pores are formed directly below UME (working electrode), with SECM it becomes possible to perform locally targeted electroporation. It has been found that in order to obtain reversible electroporation, it is necessary to leave the system for a certain time period without giving it any potential. It is also assumed that the size of the resulting pores formed should depend on the size of the UME used.

These experiments lead to a promising expectation that SECM has a great potential in controlling electroporation phenomenon.

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