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PULSED CONDUCTOMETRY OF SINGLE CELLS IN ELECTRIC FIELD WITH RISING STRENGTH

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Relevance. In biophysics, bioengineering technologies and biomedicine, various methods based on the phenomenon of electroporation of living cell membrane in a pulsed electric field (PEF) are used [1, 2]. The influence of the field leads to reversible perforation of the membrane or its irreversible rupture and lysis of the cell, depending on the PEF strength. The process and result of the interaction of the membrane and the cell with the field can be observed by nonlinear changes in their overall conductivity with gradually PEF rising strength, i.e. PEFRS. Conductivity in this case is both an indicator and an integral characteristic of the development of the electroporation process with a linear rising in the field strength up to the electrical breakdown of the membrane [3].

Purpose. This work was carried out in order to briefly present the possibilities of the pulsed conductometry method anddevice.

Results. To study the dynamics of electroporation, a method and device for pulsed conductometry of single animal cells in PEFRS were developed [2, 3]. The method allows to determine the conductivity of cells and liquid media in the range of 0.1-10^5 μS/cm in the PEFRS of 0-10 kV/cm and some biophysical parameters of the cell, for example, capacity [4]. With the help of this method it was easy to justify the necessary modes of influence on the cell of PEF for various technological applications of electroporation [2, 4, 5].

Fig.1 shows the dependences the conductivity of mouse oocyte, 2-cell embryo in 0.3M sucrose on the electric field strength.

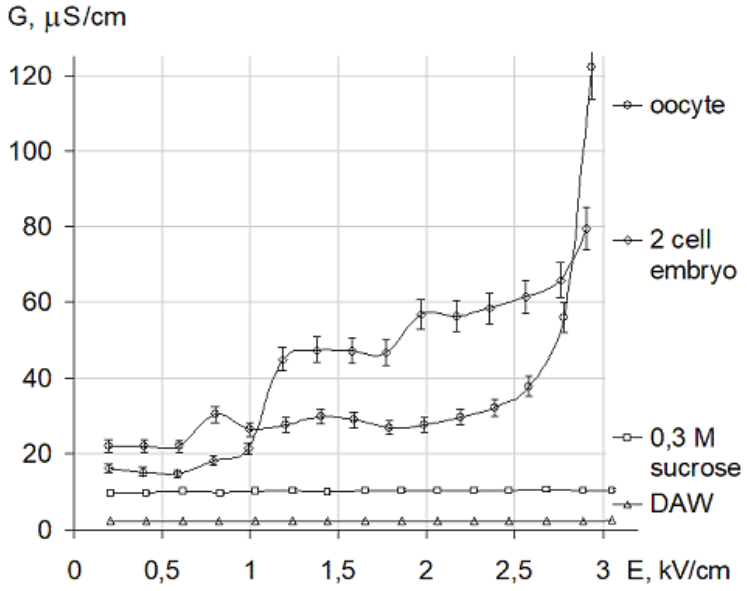

Fig. 1 – Dependences the conductivity of mouse oocyte, 2-cell embryo, deionized apyrogenic water (DAW) and 0.3 M sucrose on field strength

Curve of conductivity of the oocyte has a sharp rise – this is irreversible electric breakdown of the cell membrane at 2.6 kV/cm. Curve of conductivity 2-cell embryo shows a reversible breakdown of the contact points of the membranes of two blastomeres (electrofusion) at the field strength about 1.1 kV/cm and irreversible at 2.8 kV/cm. For both curves is visible multiple reversible electroporation of cell membranes (conductivity oscillations). The conductivity of deionized apyrogenic water (DAW) and 0.3M sucrose as dielectric media do not depend on the field

strength.

Fig. 2 shows the location of a 2-cell mouse embryo between the microelectrodes during conductivity measurements or electrofusion, and on the right is a group of embryos at various stages of fusion.

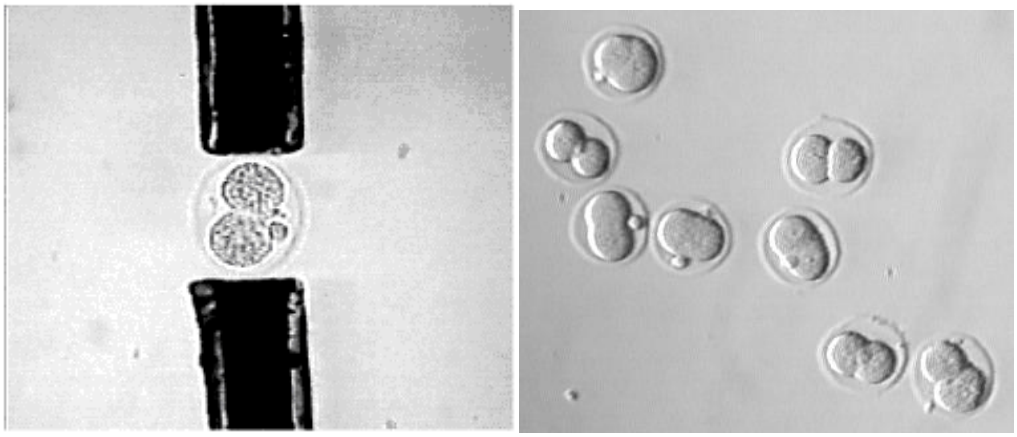

Fig. 2 – Location of 2-cell mouse embryo between the microelectrodes in the process of pulsed conductometry or electrofusion (left) and embryos at different stages of fusion (right)

Conclusions. Summarizing the results achieved, it can be concluded that the following methods and cell technologies are implemented on the basis of the method and equipment of pulsed conductometry in PEFRS [1-5]:

1. Determination of the electrical properties of membranes of different species animal cells in different liquid media for bioengineering and cryobiology.
2. Cell electrofusion (cloning, chimeras, etc.), stimulation and activation of in vitro development of oocyte-cumulus complexes and oocytes for use in reproductive biotechnology and cell engineering.
3. Ecological monitoring of the purity of natural waters and liquid foods.

There may be more other applications of the PEFRS method and device, where it is required to study the conductivity of liquid objects in a wide range of field strength in the droplet micro volume.

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