2P44

Altering Dielectric Properties of Human Cancer Cells by Varying Electrical Pulse Durations

A. L. Garner[1], J. Yang, N. Chen, J. Kolb, K. C. Loftin, R. J. Swanson, S. Beebe, R. P. Joshi, and K. H. Schoenbach

Center for Bioelectrics, Old Dominion University, Norfolk, Virginia 23510; [1]Present Address: Department of Nuclear Engineering and Radiological Sciences, University of Michigan, Ann Arbor, Michigan 48109

Microsecond-duration pulsed electric fields (PEFs) above a certain voltage cause electroporation [1], or increased permeability of the cell membrane due to pore formation, while submicrosecond pulses induce intracellular effects [2]. Models developed to describe and interpret these effects often depend on the electrical properties of the cells, which are altered by the PEF [1]. We determined the complex permittivity of a cell suspension using time domain dielectric spectroscopy (TDDS) [3]. We used a two-shell model of the cell to calculate the conductivity and permittivity of the cell membrane, cytoplasm, nuclear envelope, and nucleoplasm from the complex permittivity. For long pulses (50 μs), we found that cell membrane poration occurred within 10 s of the pulse, whereas poration was delayed by minutes for 10 ns pulses. These results indicate that membrane opening is the primary result for long pulses and a secondary result for ultrashort pulses, in agreement with other observations [4]. Membrane recovery time is similar for both pulse durations. Our initial studies have focused on temporal changes in the cell membrane. TDDS will allow us to explore electrical pulse effects on the cell nucleus.

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2P45

Evidences for Membrane Elektroporation during Application of Nanoseconds Electrical Pulses

C. Gusbeth, W. Frey, and H. Bluhm

Forschungszentrum Karlsruhe GmbH, Institute for Pulsed Power and Microwave Technology, P.O. Box 3640, D-76021 Karlsruhe, Germany

It was suggested that high intensity (>100 kV/cm) short duration (ns) electrical pulses could affect the intracellular structures of mammalian cells without adversely affecting the outer cell membrane by electroporation. In order to demonstrate this on microorganisms with and without internal structures, we have studied the effect induced by short electrical pulses (25–600ns) on membrane integrity of *Saccharomyces cerevisiae* (backer yeast) and *Pseudomonas putida* (bacteria). Cells were stained after pulse treatment with BacLight (Syto9 and propidium iodite, PI) and counted using an epifluorescence microscope. For comparison with viability studies, the cells were staining with FUN1 (yeast) CTC (bacteria) and additional plating the sample on culture media. A Blumlein generator provided square wave voltage pulses of 25 ns to the commercial (BTX) treatment chambers. Longer high voltage pulses (100–600ns) were produced using a line generator.

Viable cells, treated using short pulses, were swollen; whereas inactive cells showed almost a permeable membrane, indicated by PI. The inactivation rates of yeast and bacteria were of the same order of magnitude and comparable with those obtained by other studies. The rates of cells stained with BacLight were related to the rates obtained by counting the metabolically active cells stained with FUN1 or CTC. We found out that inactivation rates of yeast and bacteria only depend on the treatment duration ($t_{pulse} \cdot n$) and on the product of square field intensity and treatment duration ($E^2 \cdot t_{pulse} \cdot n$). These results support the assumption that the main effect of short electrical pulses (ns – range) is the electroporation of the outer cell membrane.