**Application No.** 812/21

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**Scientific abstract** –*<*Developing a reversible electroporation model of bacteria based on rate permeabilization measurements of hydrophilic and hydrophobic compounds in a moderate electric field*>*

**Scientific background:** Applying an external pulsed electric field (PEF) to microbial cells increases membrane permeability, a phenomenon termed electroporation. Drawing on theoretical studies and experiments, it has been suggested that electroporation occurs when the external electrical field exceeds the capacity of the cell membrane potential, leading to mechanical changes, including the formation of hydrophilic pores, in less than one second. Pore resealing has been reported to occur over a range of minutes or even hours. The ability of PEF application on bacterial cells to induce electroporation depends on electrical parameters, the targeted cell type, and the treatment medium. **Research goal:** To study the effect of PEF treatment on bacterial cell destruction and recovery kinetics and to develop a reversible electroporation model based on the measurement of cell permeabilization rates of hydrophilic and hydrophobic compounds in a moderate electric field. **Research design & methods**: Examine pore size and resealing time in electroporated gram-negative and gram-positive bacteria, as well as bacterial protoplast.Cells suspended in PBS will be exposed to PEF treatment and then diluted in a brain heart infusion (BHI) solution, a rich medium previously shown to promote pore resealing. The BHI will contain different compounds, such as a fluorescent dye, and hydrophobic and relative hydrophilic compounds. The permeabilization rate of each of the above compounds will be examined using flow cytometry or high-performance liquid chromatography at different time intervals until the permeabilization rate reaches zero. To further understand the mechanisms underlying PEF-treated bacteria pore resealing, we will perform proteomic analysis using mass spectrometry. Based on the experimental results, we will develop a bacterial electroporation kinetic model to analyze tracer diffusion rates into the cell, calculating the kinetics of mass transfer as well as pore destruction and recovery. Furthermore, a continuous field model will be developed to simulate the PEF exposure process. The model will be composed ofa conservation equation for mass, momentum, energy, electric potential, and the transport equation for passive biological tracer activity. The numerical tool employed isa commercially available CFD software package (COMSOL Multiphysics®), used to solve numerical 3D transient models by calculating temperature responses in space and time. **Expected significance:** The electroporation model will help predict experimental outcomes and optimize experimental protocols. Reversible electroporation of relatively small molecules will provide an important method that can be employed in multiple genetic engineering processes and lipid and protein extraction, and may also be useful for bioremediation processes.