PULSING BUFFER COMPOSITION IMPACT ON CELL VIABILITY AFTER ELECTROPORATIONBY SUPPLEMENTING THE BUFFERS WITH INTRACELLULAR MOLECULES

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Electroporation (EP) is a widely used method for intracellular delivery of biomolecules, yet the cell viability after treatment remains a critical limitation influenced by the involved pulsing conditions and respective medium composition [1]. While the extracellular environment plays a key role in gene electrotransfer optimization, its effects following membrane permeabilization have not been fully characterized [2, 3]. The objective of this study was to evaluate how EP supernatant affects cell viability post-electroporation under different pulsing media, with the aim of identifying conditions that improve cellular recovery while maintaining effective electroporation. To achieve this CHO cells were electroporated using three different media: a standard EP medium, EP HEPES buffer, and EP HEPES buffer supplemented with calcium. Following the electroporation with different parameters, cells were incubated with fresh medium or in EP-supernatant separately. Cell viability was assessed across a range of pulse strengths using flow cytometry-based analysis and MTS assay. The observed results after 24hrs represent the impact of EP-supernatant on cell viability. In the standard electroporation medium, exposure to pulses in EP-supernatant significantly improved cell viability, particularly at higher pulse strengths. In contrast, cells exposed to EP-supernatants from HEPES and HEPES supplemented with calcium showed significantly reduced viability compared to cells maintained in fresh medium. These findings clearly indicate that medium composition plays a crucial role in cellular responses post-electroporation. In conclusion, this study highlights the importance of selecting appropriate pulsing media to enhance post-electroporation cell survival [2, 3]. Ongoing work aims to assess gene electrotransfer efficiency and to investigate morphological changes associated with different media conditions, providing further insight into optimizing electroporation-based gene delivery protocols.

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