The Effect of Physical Cues on Stem Cells and Extracellular Vesicles Toward Neuropathy Applications

Abstract

Extracellular vesicles (EVs) derived from human mesenchymal stem cells (hMSCs) and Schwann cells (hSCs) present a promising cell-free therapeutic strategy for peripheral nerve regeneration. However, their clinical translation is severely bottlenecked by low baseline cellular secretion rates, batch-to-batch heterogeneity, and a lack of scalable bioprocessing frameworks. Traditional strategies to boost yield often rely on mechanical stress, genetic manipulation, or chemical stimulants that compromise cell viability and cargo consistency. To resolve these biomanufacturing limitations, this dissertation investigated a series of biophysical hydrodynamic, and material-based surface engineering strategies designed to amplify EV output while preserving stem cell health, metabolic stability, and therapeutic potency.

Across these bioprocessing platforms, physical and material-based cues were shown to directly regulate EV biogenesis and cargo composition. Low-frequency direct current electrical stimulation (ES) served as an effective biophysical trigger, significantly increasing EV secretion from human adipose derived mesenchymal stem cells (hASCs) and hSCs through the targeted upregulation of endosomal sorting complex (ESCRT)-dependent and ESCRT-independent biogenesis pathways while modulating neuroinflammatory cytokine expression. To transition these findings into scalable dynamic architectures, 3D hBM-MSC expansion within a Vertical-Wheel bioreactor (VWBR) leveraged fluid shear stress to drive metabolic shifts toward glycolysis, achieving a 3- to 10-fold increase in EV yield over static controls while enriching therapeutic microRNAs (including miR-29a-3p and miR-143-3p) capable of suppressing pro-inflammatory markers in activated hSCs. Finally, to decouple yield amplification from cellular stress, microcarriers engineered with tannic acid-iron (TA-Fe) and calcium-supplemented (TA-Fe-Ca) metal-phenolic network (MPN) coatings optimized interfacial bioactivity, supporting sustained expansion and substantially boosting EV output through the selective transcriptional activation of ESCRT machinery without elevating baseline metabolic energy demands.

Collectively, this body of work demonstrates that biophysical, hydrodynamic, and surface-engineering strategies can independently and synergistically overcome production bottlenecks in cell-free biomanufacturing. By directing vesicle biogenesis mechanisms at the cell environment interface without inducing cellular exhaustion, these platforms provide a scalable, economically viable foundation to accelerate the clinical translation of therapeutic EVs.