FAMU-FSU College of Engineering

The Joint College *for* Florida A&M Univeristy
and Florida State University

Department of Chemical and Biomedical Engineering

Dissertation Defense

DEPARTMENT OF CHEMICAL & BIOMEDICAL ENGINEERING | FAMU-FSU College of Engineering

By: Justice Ene (Advisor: Yan Li)
Date: Wednesday, September 02, 2026
Time: 2:00pm - 4:00pm
Location: Virtual Zoom Meeting - https://fsu.zoom.us/j/96081223773 | In-person, IRCB 1030

Engineering Extracellular Vesicles of Lineage Specific Three-Dimensional Organoids Derived from Human Pluripotent Stem Cells for Treating Senescence and Neural Degeneration

Abstract

Stem cell therapy has emerged as a promising approach in regenerative medicine, particularly through the use of organoids derived from induced pluripotent stem cells (iPSCs), which are adult cells reprogrammed into a pluripotent state capable of differentiating into nearly any cell type. While iPSC-based therapies have advanced models for neurodegenerative diseases such as Alzheimer's disease (AD) and age-related cognitive decline, challenges including tumor formation, immune rejection, and safety concerns limit their clinical application. Extracellular vesicles (EVs), membrane-bound nanoparticles secreted by cells and enriched with proteins, lipids, and nucleic acids, offer a promising cell-free alternative. Stem cell-derived EVs have demonstrated therapeutic potential through anti-inflammatory, antioxidant, wound-healing, and anti-senescent effects. A major obstacle to clinical translation is the limited yield of EVs produced using conventional static culture systems. This study investigated Vertical-Wheel Bioreactors (VWBRs) as a scalable platform for producing EVs from human iPSC-derived blood vessel organoids (BVOs). BVOs were cultured as aggregates or on microcarriers under varying shear stress conditions. Low-shear VWBR cultures produced 2–3-fold higher EV yields per milliliter than static controls. Dynamic culture also altered EV lipid composition, demonstrating that bioprocess conditions influence EV cargo and characteristics. In particular, VWBR-derived EVs retained therapeutic efficacy by reducing cellular senescence in vitro. The second phase of this work evaluated EV therapeutic potential in increasingly complex in vitro models of aging and neurodegeneration. Senescence was induced using D-galactose and conditioned media from AD patient-derived brain organoids containing disease-associated cytokines, EVs, and secreted factors. These models more closely replicated the biochemical environment of neurodegenerative disease. Therapeutic outcomes demonstrated that the differentiation state of the parental cells, EV dosage, and cargo loading strategies (e.g., miR-124 loaded EVs) significantly influenced the anti-aging effects of organoid-derived EVs, supporting their potential as customizable therapeutic delivery vehicles capable of crossing the blood-brain barrier. Finally, this study examined how AD genetic background influences EV cargo and function and the effects of healthy organoid EVs on reducing neurodegeneration. EVs secreted by human forebrain organoids carrying sporadic (APOE4) or familial (APP/Presenilin-1) AD mutations displayed distinct lipidomic, proteomic, and miRNA profiles compared with healthy controls. Multi-omics analyses identified dysregulated pathways associated with ferroptosis, oxidative stress, and neurodegeneration. AD-derived EVs increased reactive oxygen species and reduced mitochondrial activity in recipient cells, whereas EVs from healthy donor cells mitigated these pathological changes. Co-culture studies with microglia further demonstrated cell type-specific interactions and the therapeutic benefits of healthy brain organoid EVs. Together, these findings establish scalable EV biomanufacturing methods, identify factors governing therapeutic efficacy, and highlight healthy brain organoid-derived EVs as promising cell-free therapeutics for Alzheimer's disease and age-related neurodegeneration

Key words: biomanufacturing, human pluripotent stem cells, extracellular vesicles, neurodegeneration, Alzheimer's disease, APOE4