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An approach to reconstructing an outer retina from human embryonic stem cells to study pharmaceuticals and mechanisms of retinal degeneration

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Retinal pigment epithelium (RPE) and retinal progenitor cells (RPC) can be derived from human embryonic stem cells (hESC). These preparations have been studied in vitro and can to a remarkable degree. A shortcoming of using monocultures to understand the behavior of tissues for differentiate grafting, or the effects of pharmaceuticals, is that the RPE forms the outer blood-retinal barrier and functions intimately with photoreceptors. To study the effects of these tissue interactions an experimental model has to satisfy two conditions: a culture medium is required that is compatible with each tissue and a scaffold must allow the tissues to interact with an in vivo-like geometry. RPE and RPC were derived from two hESC lines. The hESC-RPE was compared to human fetal RPE (hfRPE) that was isolated from 16-week fetuses. All cultures were adapted to the serum-free medium. Electrophysiologic studies showed RPE barrier properties, such as permeability and selectivity, were similar in hESC-RPE and hfRPE. RNA sequencing demonstrated that the transcriptomes of hESC-RPE and hfRPE were quantitatively similar, but there significant differences in the expression of adhesion junction and membrane transport genes. These were confirmed by quantitative RT-PCR, immunoblotting and immunofluorescence. The RPC could be differentiated and maintained on sheets of electrospun fibers of polycaprolactone. Ongoing experiments indicate that gene expression in RPE and RPC becomes more mature in co-culture. These data indicated that hESC-derived RPE is highly differentiated, but appears less mature than RPE isolated from 16-week fetuses.

Biography

Rizzolo completed his Ph.D. at the age of 26 from Duke University and performed postdoctoral studies at Harvard Medical School and New York University School of Medicine. He is an Associate Professor of Surgery with a joint appointment in Ophthalmology and Visual Science at Yale University, where he won the Bohmfalk Prize for teaching. His research focuses on the outer blood-retinal barrier and degenerative diseases of the retina. He is a Fellow of the Association for Research in Vision and Ophthalmology. He has published more than 50 peer-reviewed papers and serves on numerous Editorial Boards and grant review panels.

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