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Evaluating Estradiol as a Novel Therapy for Dry Age-related Macular Degeneration/Geographic Atrophy

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PURPOSE:

Age-related macular degeneration (AMD) is the most common cause of irreversible vision loss in the elderly population in the United States and is more prevalent in post-menopausal females than age-matched males. There are no current FDA-approved therapies for dry AMD, which composes 80-90% of all AMD cases. Late stage dry AMD (geographic atrophy) is characterized by extensive loss of the retinal pigment epithelium (RPE) along with degeneration of the photoreceptors, which cause significant central vision loss. AMD is characterized by drusen, which are extracellular deposits located beneath the retina. Drusen contain proteins associated with the process of inflammation such as activated complement proteins, fragments of the membrane attack complex, vitronectin, cholesterol, C-reactive protein, and amyloid-β. Amyloid-β is a cytotoxic protein associated with various neurodegenerative diseases and is commonly found in plaques of Alzheimer's disease.

Based on these findings, we hypothesized that estradiol may have protective effects against the cytotoxic properties of amyloid-b in RPE cells *in vitro*.

METHODS:

ARPE-19 cells were pretreated with 17-β estradiol 20-160 nM for 6 h and then stressed with 5 μM active amyloid-β_{1-42} for 24 h. Scrambled amyloid-β_{42-1} and equivalent volume DMSO solutions were used as negative controls. Cell viability (MTT Assay), reactive oxygen species (ROS) production (H_2DCFDA Assay), and mitochondrial membrane potential (JC-1 Assay) were assessed. Statistical analysis was performed by GraphPad Prism software. Statistical significance was determined at P42-1.

RESULTS:

ARPE-19 cells stressed with 5 μM amyloid-β_{1-42} had a significant decrease in cell viability (P = 0.0007), which was not reversed by pretreatment with 20 and 40 nM estradiol. The 5 μM active amyloid-β_{1-42} did not alter the levels of ROS or mitochondrial membrane potential compared to the scrambled amyloid-b_{42-1}. In addition, pretreatment with 20-160 nM estradiol did not significantly decrease ROS production or increase mitochondrial membrane potential in amyloid-b_{1-42} treated cultures.

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CONCLUSION: Amyloid-$\beta_{1\text{-}42}$ decreases ARPE-19 cell viability but not through mechanisms of ROS production or by affecting the mitochondrial membrane potential. Although estradiol has been shown to be protective on the retina when stressed with light or high glucose-induced damage, our findings demonstrate that with amyloid-$\beta_{1\text{-}42}$-induced damage, the estradiol was unable to prevent RPE death

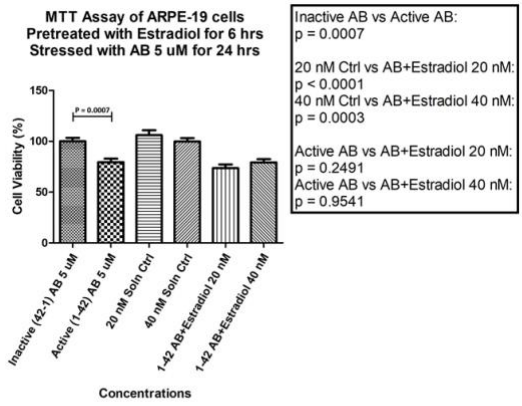

Figure 1. MTT Assay for Cell Viability. Amyloid-β 5 uM ± Estradiol 20-40 nM.

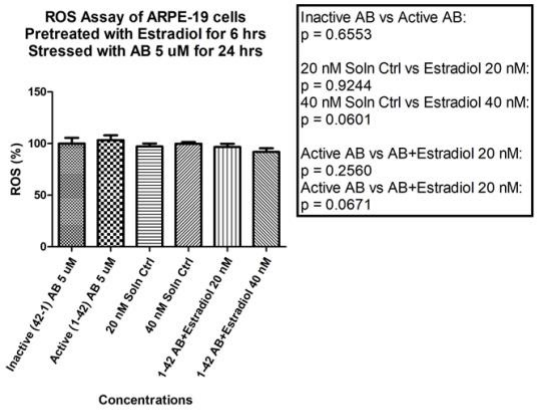

Figure 2. ROS Assay for Reactive Oxygen Species. Amyloid-β 5 uM ± Estradiol 20-40 nM.

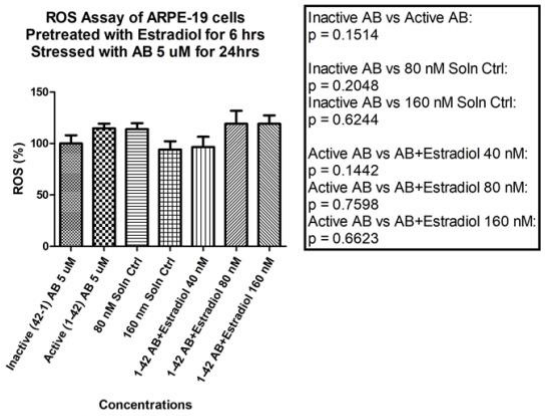

Figure 3. ROS Assay for Reactive Oxygen Species. Amyloid-β 5 uM ± Estradiol 40-160 nM.

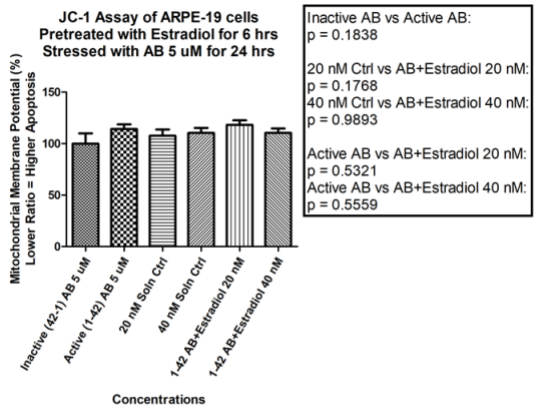

Figure 4. JC-1 Assay for Mitochondrial Membrane Potential. Amyloid-β 5 uM ± Estradiol 20-40 nM.

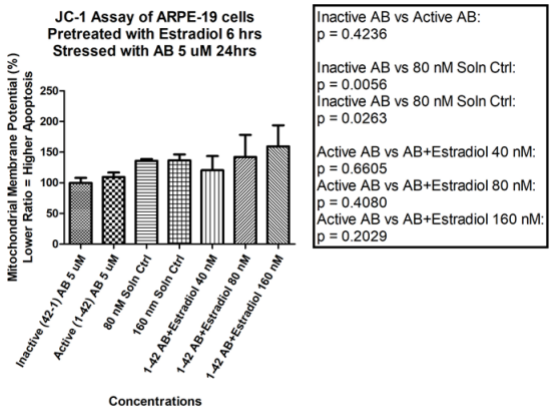

Figure 5. JC-1 Assay for Mitochondrial Membrane Potential. Amyloid-β 5 uM ± Estradiol 40-160 nM.