**Reversible Mitochondrial Injury in Dying Retinal Ganglion Cells**

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**Abstract**

**Purpose:** Glaucoma is a neurodegenerative disease in which various triggers induce cascades of secondary events, which ultimately lead to retinal ganglion cell (RGC) death. Programed cell death is generally considered to be irreversible. However, recent studies reveal that recovery of dying cells is possible, even after reaching critical cell death events. This phenomenon is termed anastasis. Harnessing mechanisms of anastasis may represent a previously unrecognized therapeutic strategy to rescue dying differentiated cells that are difficult to replace. We study neuronal cell death with the aim of rescuing dying RGCs in glaucoma.

**Methods:** Primary ratRGCs anddifferentiated PC12 cells were treated with ethanol to induce cell death. Live cell imaging with fluorescent probes (Mito-Tracker, TMRM, DCFDA, Fluo-8AM) were used to visualize cell injuries in individual RGCs and differentiated PC12 cells with high-resolution live-cell spinning disk confocal microscopy. Electron microscopy (EM) was used to observe the ultrastructure of mitochondria.

**Results:** The results showed that RGCs and differentiated PC12 cells could recover from mitochondrial fragmentation and membrane potential loss by removing the cell death stimulus ethanol and further culturing in fresh cell culture medium. EM results confirmed the ethanol induced mitochondrial fragmentation and its reversibility. Furthermore, we observed that reversible mitochondrial fragmentation was accompanied with reactive oxygen species generation and intracellular Ca2+ elevation. However, no cytochrome c release from mitochondria was observed at this stage.

**Conclusion:** The results indicated that targeting fragmented or dysfunctional mitochondria may be an effective therapeutic strategy to rescue dying RGCs and reduce the loss of vision in glaucoma.

Key words: Retinal ganglion cells; cell death; mitochondria fragmentation