Metabolic mapping of photoreceptor degeneration

Ferrell WD, Vázquez-Chona FR, Jones BW & Marc RE
Moran Eye Center University of Utah

Photoreceptors function in an oxidatively challenging environment and are vulnerable to metabolic and energetic imbalances. Yet, in-depth profiling of photoreceptor metabolism during the degenerative process remains incomplete. We relate the classic sequence of photoreceptor degeneration and glial activation to small molecular metabolic signals using computational molecular profiling (CMP).

**CMP explores metabolite concentration with high spatial resolution and high dimentionality**

Light-induced oxidative damage induces metabolic diversity within the photoreceptor population

**Photoreceptor degeneration sequela is parsable into quantifiable metabolic profiles**

Histograms showing number of pixels vs pixel intensity:

Molecular profiles were derived using k-means algorithm. Each profile can produce a histogram for their respective metabolite or proteomic signal. Stacked bivariate histograms describing metabolic diversity within the photoreceptor cohort. Each plot represent eight signals mapped on a y-concentration coordinates. Each plot spans estimated concentration range 0.1-10 mM.

Glutamate depletion correlates with DNA fragmentation

Fluorescent TUNEL labelling in green with Glutamate staining in magenta in LR white resin. Arrows indicate TUNEL+/Glutamate+ cells. Asterisks indicate TUNEL+/Glutamate+ cells. These findings suggest that TUNEL staining is a late DNA indication of cell death events that are more acutely represented in the metabolite.

**Surviving cones & rods as well as degenerating & apoptotic cells display unique metabolic profiles**

Chromatin morphology

Class Region ultrastructure signature
(1) pink Endfeet Ribosomes Intermediate filament vacuoles High: GS, E, Q, TT, CRALBP Moderate: J, R Low: Methionine (M)
(2) blue IPL. Trunk Ribosomes Intermediate filament Medium: GS, TT, R Low: M, GABA
(3) green near dying cells cytoplasm + vacuoles Intermediate filament High: Q, CRALBP Moderate: GS, E, TT, J Low: R
(4) red around stressed cells nucleus Thick interm. filament bundles Chromatin Medium: Q High: Medium: E, TT, J, CRALBP Low: GS, R

Visualization of metabolic diversity can help predict mechanisms of retinal degeneration and glial activation

**Na+/K+ ATPase collapse may decrease ion gradients and lead to reverse transport of Glutamate & Taurine**

Ultrasound and metabolic analyses of albino BALB/c retina after 24 h light damage. EM mosaic represents nearly 200 tiles at 5000x. Taurine-Glutamine-Glutamate (RGB) composite

Corroborating evidence:

- Sodium channel blockers reduce glutamate release (Roy et al., 1995)
- Increasing extracellular K+ concentration increases glutamate release (Goskowik et al., 1996)

**Müller glia are highly sensitive to neuronal stress and respond to stress events with focal small molecular responses**

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