

# Distortion of axonal cytoskeleton: an early sign of glaucomatous damage

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**PURPOSE:** Glaucoma damages the retinal nerve fiber layer (RNFL) . The purpose of this study was to investigate the distribution of major cytoskeleton components, F-actin, microtubules (MTs) , and neurofilaments (NFs) , in the RNFL during the development of glaucoma.

**METHODS:** Intraocular hypertension was induced in a rat model by laser photocoagulation of the trabecular meshwork. Retinas were obtained after 2 to 3.5 weeks of treatment. Multiple fluorescent stains were used to identify F-actin, MTs, NFs, and nuclei simultaneously in the same tissue. Distribution of these components in a whole-mounted retina was examined by confocal microscopy. Fluorescent stain was quantitatively described.

**RESULTS:** In normal RNFL F-actin, MTs, and NFs were intensely stained. Along the bundles, F-actin and MTs were strongly colocalized, but alternating strands of F-actin and NFs were apparent. Normal RNFL lacked nuclei. In glaucomatous retinas, irregular staining of F-actin, MTs, and NFs was found within the bundles. A strong network of F-actin appeared on the RNFL surface and between the bundles. In severely damaged retinal regions total loss of F-actin and MTs was found, whereas residual strands of NFs were evident. Before the decrease in RNFL thickness, irregularity of F-actin stain and density of nuclei in the RNFL significantly increased.

**CONCLUSIONS:** The results suggest that F-actin, MTs, and NFs are rich and approximately uniformly distributed in the normal RNFL. Glaucoma causes alteration of the cytoskeleton in the RNFL. F-actin is the most sensitive component in its response to stress on the retina. An increase in the number of nuclei in the RNFL may be an early sign of glaucomatous damage.

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