Abstract Information

Abstract Title:
Transgenic Studies on the Role of Myocilin and Optineurin in the Eye

Purpose:
To obtain experimental in vivo information on the functional properties of myocilin and optineurin for aqueous humor outflow and to study in vivo the processing of mutated Tyr437His myocilin. Mutations in myocilin and optineurin are responsible for some forms of primary open-angle glaucoma, and patients with the Tyr437His mutation have severe phenotypes.

Design:
N/A

Participants:
N/A

Main Outcome Measures:
N/A

Methods:
The chicken betaB1-crystallin promoter was used to overexpress optineurin, wildtype human myocilin, and Tyr437His myocilin in the lenses of transgenic mice. Expression of transgenic mRNA was monitored by northern blot analysis and in situ hybridization. The localization and secretion of transgenic proteins was investigated by western blot analysis, and light and electron microscopy. Intraocular pressure (IOP) was measured by anterior chamber cannulation.

Results:
Two independent lines were established with each of the constructs that showed a strong expression of transgenic mRNA in their lenses. Transgenic expression resulted in a 4.7 ±1.8 increase of secreted myocilin in mouse aqueous humor, compared with its concentration in human aqueous humor. Immunoreactivity for transgenic myocilin was observed
along the surfaces of lens and corneal endothelium, and in the chamber angle. At 12 weeks of age, the ultrastructure of the trabecular meshwork in mice expressing normal myocilin was not different from that of control eyes and IOP did not significantly differ from that of control littermates. In contrast, mutated Tyr437His was not secreted from lens fibers, but accumulated in dilated cisterns of rough endoplasmic reticulum. While no structural changes were observed in lenses of animals expressing normal myocilin, lenses with Tyr437His expression developed nuclear cataracts, completely lost transparence and eventually ruptured. Immunohistochemistry showed a broad cytoplasmic distribution of optineurin in lens fibers. No secreted optineurin could be detected in the aqueous humor of transgenic mice or wildtype littermates.

**Conclusion:**
Our results do not support the concept that increasing amounts of myocilin in the outflow tissues obstruct the system and directly cause an increase in outflow resistance. Mutated Tyr437His myocilin is not secreted in vivo and causes severe alterations of cellular structure and function. Transgenic in vivo data indicate that optineurin is a cytoplasmic protein, which is not secreted.