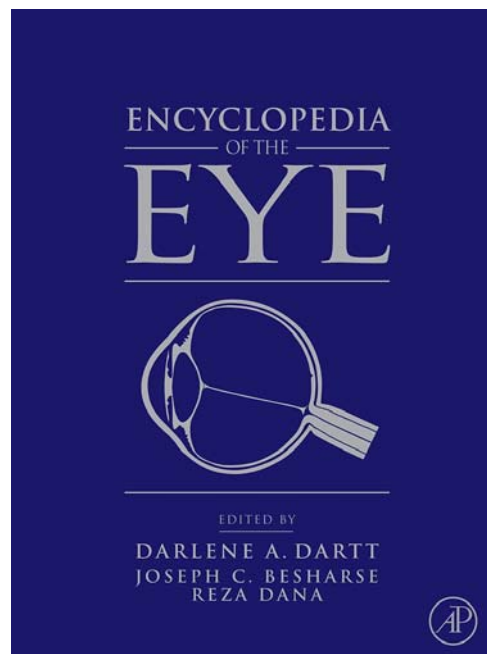


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The Cytoskeletal Network of the Trabecular Meshwork*

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Glossary

Actomyosin – The contractile cytoskeletal system, consisting of actin and myosin that, together with additional regulatory components, constitutes a force-generating system in muscle and nonmuscle cells.

Adherens junctions – The cell–cell adhesion sites (commonly referred to as: junctions) that are associated with the actin cytoskeleton, via cadherin receptors.

ATP – Adenosine 5'-triphosphate (ATP) is a multifunctional nucleotide that is most important as a molecular currency of intracellular energy transfer. In the context of this article, ATP plays a major role in cytoskeletal contraction.

Cytoskeleton – The internal network of fibers present within the cell's cytoplasm, composed largely of actin filaments, intermediate filaments, and microtubules.

Focal adhesions – Also known as focal contacts, these are specific types of large, membrane-bound macromolecular assemblies through which cells attach to the extracellular matrix (ECM). They are associated with the actin cytoskeleton, and their adhesion to the ECM is mediated by integrin receptors.

Glaucoma – A group of eye diseases characterized by a specific loss of the retinal nerve fiber layer, excavation of the optic disk, and visual-field deficits, typically caused by an intolerance to elevated or even normal intraocular pressure, and potentially leading to blindness.

Phosphorylation – The addition of a phosphate group to an organic molecule, thereby affecting the biological properties of the phosphorylated molecule. For example, phosphorylation of protein molecules (by enzymes, known as protein kinases) can affect their overall structure, interactions with other molecules, and biological activity.

Polymerization – A chemical process whereby individual components (defined as monomers) interact with each other, forming a stable molecular chain.

Schlemm's canal – A circular endothelium-lined channel that is located in the front of the eye internal to the limbus. It collects aqueous humor from the anterior chamber and drains it into the general circulation.

Trabecular meshwork – An area of tissue composed of arrays of collagen beams covered by endothelial-like cells with ECM occupying the spaces. It is located in the angle of the anterior chamber internal to Schlemm's canal and allows aqueous humor to drain into Schlemm's canal from the anterior chamber.

Actomyosin System in the Trabecular Outflow Route

Aqueous humor enters the posterior chamber of the eye from the ciliary processes, flows around the lens and through the pupil into the anterior chamber, and leaves the eye primarily through the trabecular route and the uveoscleral route at the anterior chamber angle. The trabecular route is the predominant outflow pathway in human eyes and consists of the trabecular meshwork (TM) and Schlemm's canal. The TM is composed of arrays of collagen beams covered by endothelial-like cells, with loose extracellular matrix (ECM) occupying the spaces between the cells of the adjacent beams. The outermost, juxtacanalicular (JCT) or cribriform region has no collagenous beams, but rather several cell layers immersed in a loose web of ECM fibrils. The adjacent Schlemm's canal is a continuous endothelium-lined channel that drains aqueous humor to the general venous circulation. TM structure and experimental flow studies suggest that flow resistance is maximal in the JCT region and/or the inner wall of Schlemm's canal, although the exact location of the major resistance barrier is not clear. Glaucoma is an ophthalmologic disorder responsible for visual impairment. Generally, glaucoma is characterized by progressive optic neuropathy usually associated with elevated or intolerable intraocular pressure (IOP), consequent to abnormally high flow resistance in the TM. Since glaucomatous eyes exhibit fewer TM cells and abnormally appearing JCT ECM compared to the eyes of age-matched normal individuals, cells and ECM in the JCT region may be critical in resistance regulation. In the last two decades, dynamics of the actin cytoskeleton in TM/Schlemm's canal cells have been confirmed to play important roles in the regulation of aqueous humor outflow.

*An adaptation and extension of Tian, B., Gabelt, B. T., Geiger, B., and Kaufman, P. L. (2009). The role of the actomyosin system in regulating trabecular fluid outflow. *Experimental Eye Research* 88: 713–717. Copyright Elsevier (2008).

The actomyosin system, composed of actin microfilaments and associated proteins, is one of the three major systems (the actomyosin system, the microtubule system, and the intermediate filament system) of the cytoskeleton. Microfilaments assemble within cells into complex bundles or three-dimensional meshworks located subjacent to the plasma membrane, and attach to the plasma membrane at a variety of sites, including adherens cell–cell junctions and focal adhesions, through specific receptors, namely cadherins and integrins, respectively. This link is mediated through a network of anchor proteins and additional structural and signaling molecules. The actomyosin system is present in essentially all cells, including TM and Schlemm's canal cells. Microfilament-based structures, such as adherens cell–cell junctions, focal contacts, and microfilament bundles, are highly organized in the cells along the trabecular outflow pathway (Figure 1). Intact microfilament bundles in TM/Schlemm's canal cells are essential to maintain physiological contractility of the JCT–Schlemm's canal region. Conversely, a physiologically contracted state of the JCT–Schlemm's canal region is required to maintain the microfilament-related structures in the outflow pathway. Microfilaments are involved in a variety of cellular processes from cell adhesion and motility to organelle trafficking to adhesion-mediated signal transduction. Therefore, microfilament dynamics play important roles in cellular morphogenesis, such as changes in cell shape, volume, contractility, and adhesion to neighboring cells and to the ECM. These changes in TM and/or Schlemm's canal cells, which could affect trabecular outflow resistance by altering the dimensions or direction of flow pathways, the amount and composition of the ECM, and the structures of adherens cell–cell junctions and focal adhesions, can be modulated directly by actin-disrupting agents or indirectly by inhibition of specific protein kinase(s) or cellular contractility through administration of protein kinase inhibitors or gene therapies. In addition, since the three cytoskeleton systems function in concert, changes in the microtubule system or intermediate filament system can also induce radical changes in the actomyosin system itself.

Trabecular Outflow Enhancement Following Actomyosin Inhibition

Disruption of Microfilaments

The actin microfilament (filamentous actin; F-actin), which is the major component of the actomyosin system, is composed of many G-actin (globular actin) monomers. Pharmacological disruption of microfilaments in TM and Schlemm's canal cells alters the cell shape, inhibits cellular contractility, affects adhesions of cell–cell and cell–ECM, and in turn decreases outflow resistance in the trabecular outflow pathway.

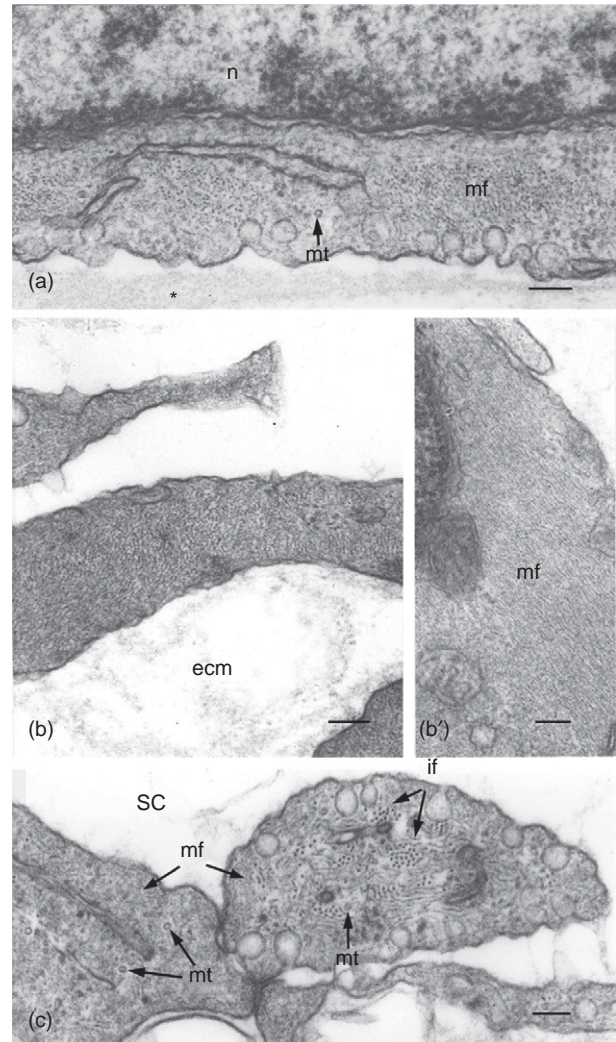


Figure 1 Transmission electron microscopy showing abundance of cytoskeletal filaments in different cellular compartments of the trabecular outflow pathway in a monkey eye. (a) Ventral aspect of a TM cell, which interacts with underlying collagen beam (*) through many focal-adhesion-like structures, is highly enriched with cytoskeletal filaments, including actin-rich microfilaments (mf) and microtubules (mt); n, nucleus. (b, b') Juxtacanalicular cells displaying numerous microfilaments (mf), seen in cross section (b) or longitudinal (b') section; ECM, extracellular matrix. (c) Inner-wall endothelial cells of Schlemm's canal (SC), displaying numerous intermediate filaments, organized in discrete bundles (if). Microtubules (mt) are also present. Microfilaments (mf) are usually seen in the cortical cytoplasm and near cell–cell junctions. Magnification = 0.1 μ m. from Tian, B., Geiger, B., Epstein, D. L., and Kaufman, P. L. (2000). Cytoskeletal involvement in the regulation of aqueous humor outflow. *Investigative Ophthalmology and Visual Science* 41: 619–623. Reprinted with permission of ARVO.

Cytochalasins

Cytochalasins are fungal metabolites that interfere with the polymerization process by which G-actin aggregates into F-actin. Anterior chamber infusion of microgram to milligram doses of cytochalasins B or D, in live monkey

eyes and/or organ cultures of enucleated postmortem human eyes, cause distension of the cribriform meshwork, separation of its cells, and ruptures of the inner-wall endothelium of Schlemm's canal, leading to washout of ECM and significant increases in trabecular outflow facility. This is completely independent of ciliary muscle contraction, because the effect persists in the ciliary muscle-disinserted live monkey eye and in the cultured human anterior segment in which the ciliary muscle is not functionally interacting with the TM. Subthreshold doses of cytochalasin B and the trabecular outflow enhancer H-7 (see below) significantly increased outflow facility in living monkeys, indicating that the pathways by which the two drugs reduce aqueous humor outflow resistance converge at some point presumably involving deterioration of actin microfilaments in TM/Schlemm's canal cells. *In vitro* studies with cultured human and monkey trabecular cells confirm the alteration of cell shape and, with human cells grown on filters, the increased hydraulic conductivity.

Latrunculins

Latrunculins, which are macrolides produced by the marine sponge *Negombata magnifica*, are specific and potent actin-disrupting agents. They sequester monomeric G-actin, leading to massive disassembly of the polymeric form, namely F-actin. The addition of latrunculin A or B causes destruction of microfilament bundles and associated proteins in a wide variety of cultured cells, including human TM cells. This effect is manifested by cell rounding and retraction of the lamellipodium, and is accompanied by an apparent arborization of the cells. In living monkey eyes or organ cultures of enucleated porcine or postmortem human eyes, latrunculin A or B induces major increases in outflow facility and/or decreases IOP. Morphological studies indicate that the latrunculin-B-induced decrease in outflow resistance is associated with microfilament-disruption-related structural changes in the TM. Electron microscopy of the live monkey eye has revealed substantial ballooning of the JCT region following latrunculin B treatment, leading to a substantial expansion of the space between the inner wall of Schlemm's canal and the trabecular collagen beams without observable separations between inner-wall cells (Figure 2). In postmortem human eyes, the facility increase is accompanied by increased openings between inner-wall cells (more border or paracellular pores) with only very modest rarefaction of the JCT tissue and separation of the inner wall of Schlemm's canal from JCT tissue.

Swinholide A

Unlike latrunculins, swinholide A, another marine macro-lide, severs microfilaments but stabilizes the dimeric form. However, swinholide A significantly increases outflow facility in living monkeys similar to latrunculins. Since

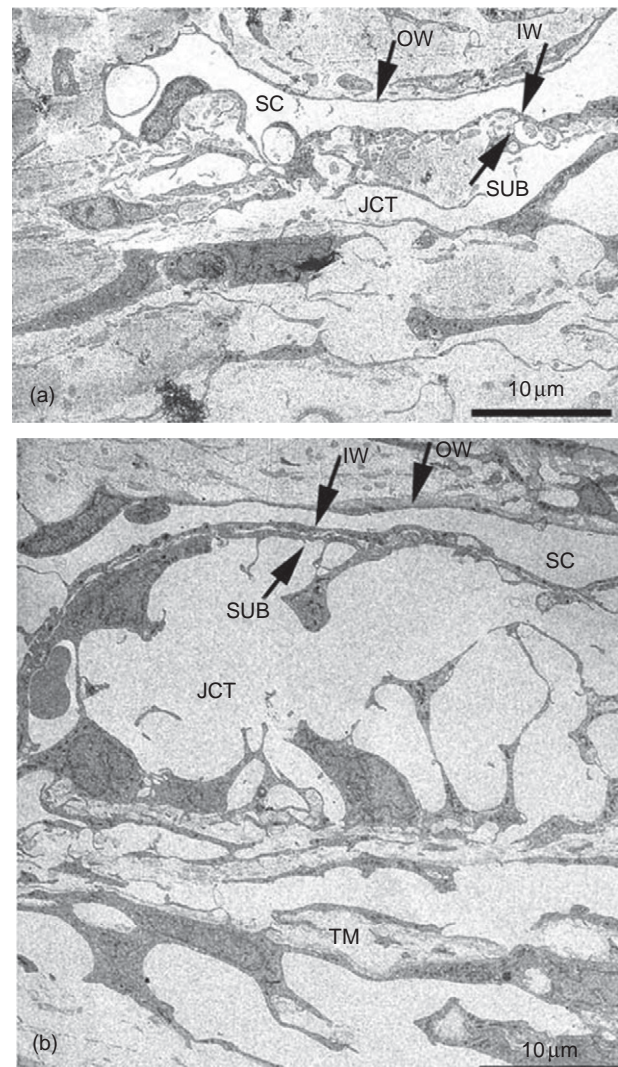


Figure 2 Transmission electron microscopy of the trabecular meshwork (TM) of a monkey eye following vehicle or LAT-B treatment: (a) shows normal JCT region and its circumjacent structures (vehicle-treated eye); (b) indicates the massive ballooning of the JCT region and the retention of close contact between IW and SUB (LAT-B-treated eye). IW, inner wall; JCT, juxtacanalicular region; OW, outer wall; SC, Schlemm's canal; SUB, subcanalicular cells. Modified from Sabanay, I., Tian, B., Gabelt, B. T., Geiger, B., and Kaufman, P. L. (2006). Latrunculin B effects on trabecular meshwork and corneal endothelial morphology in monkeys. *Experimental Eye Research* 82: 236–246. Copyright Elsevier.

swinholide A decreases the level of F-actin without significantly increasing the concentration of G-actin, its positive effect on outflow facility further indicates that microfilament depolymerization or consequent disorganization of the actomyosin system in the TM/Schlemm's canal, and not the increase in G-actin concentration, is the major mechanism responsible for the latrunculin-induced increase in outflow facility.

TM Contractility Inhibition

Muscle and nonmuscle cellular contraction is associated with Ca^{2+} -dependent activation of myosin light-chain kinase (MLCK) and consequently phosphorylation of the regulatory myosin light chain. This phosphorylation, and consequent contraction, can be greatly enhanced by G-protein-mediated activation, in which the small G protein known as Rho activates a protein kinase, namely Rho kinase (ROCK). Rho, as well as other G proteins, including Rac and Cdc42 (signaling proteins that are activated by guanosine triphosphate (GTP), thereby regulating the activity of different partner proteins which, in turn, regulate the organization of the actin cytoskeleton) play key roles in the regulation of cellular contraction. Rho-activated Rho kinase triggers myosin II activity by inhibiting myosin light-chain phosphatase as well as by phosphorylating the myosin II regulatory light chain. Myosin II is the major cytoskeletal protein that drives the assembly of contractile bundles of actin and myosin, known as stress fibers in nonmuscle cells and is responsible for the generation of cellular tension. The TM possesses smooth-muscle-like properties and the actomyosin system in TM cells plays a key role in its contraction. Expression of nonmuscle myosin IIA and IIB has been confirmed in human TM cells, suggesting that myosin II activity is also involved in the actomyosin-driven TM contractility. Pharmacological inhibition of TM contractility enhances trabecular outflow facility in living animals and cultured anterior segments of enucleated animal or human eyes.

Broad specificity protein kinase inhibitors (H-7)

H-7, a broad spectrum serine–threonine kinase inhibitor, dramatically inhibits actomyosin-driven contractility. This leads to cellular relaxation, deterioration of the microfilaments and perturbation of their membrane anchorage, and loss of stress fibers and focal contacts in many types of cultured cells. H-7 increases outflow facility and decreases IOP in living monkeys and/or organ cultures of enucleated

porcine, monkey, or postmortem human eyes, similar to actin disruptors. Morphological studies in the live monkey eye indicate that the H-7-induced increase in outflow facility is associated with cellular relaxation and drainage-surface expansion of the TM and Schlemm's canal, accompanied by loss of ECM. The inner-wall cells of Schlemm's canal become highly extended, yet cell–cell junctions are maintained (**Figure 3**). The morphological changes in the TM of live monkey eyes are consistent with functional changes in isolated bovine TM strips, where the TM precontracted by carbachol was relaxed by H-7. In post-mortem cultured anterior segments of human eyes, H-7 causes a partial loss of the endothelial lining of Schlemm's canal.

The specific target kinases affected by H-7 are not well defined, because H-7 inhibits multiple protein kinases, including MLCK, Rho kinase, and protein kinase C (PKC). Studies have confirmed that the nonselective PKC inhibitor staurosporine, the specific PKC inhibitors chelerythrine and GF109203X, and the specific MLCK inhibitor ML-7 similarly increase outflow facility in living monkeys or cultured porcine anterior segments.

Rho-kinase inhibitors (Y-27632, Y39983, HA-1077, H-1152, and INS117548)

Inhibition of Rho kinase may play a key role in regulating trabecular outflow. A specific Rho kinase inhibitor, Y-27632, induces reversible changes in cell shape and decreases in actin stress fibers, focal adhesions, and protein phosphotyrosine staining in human TM cells and Schlemm's canal cells. In isolated bovine TM strips, Y-27632 completely blocks Ca^{2+} -independent phorbol myristate acetate or endothelin-1-induced contraction. As expected, Y-27632 and other Rho kinase inhibitors (such as Y-39983, HA-1077, H-1152, and INS117548) increase outflow facility and/or decrease IOP in living rabbits, rats, monkeys, and/or enucleated porcine eyes, similar to H-7. A recent morphological study in bovine eyes indicates that, with

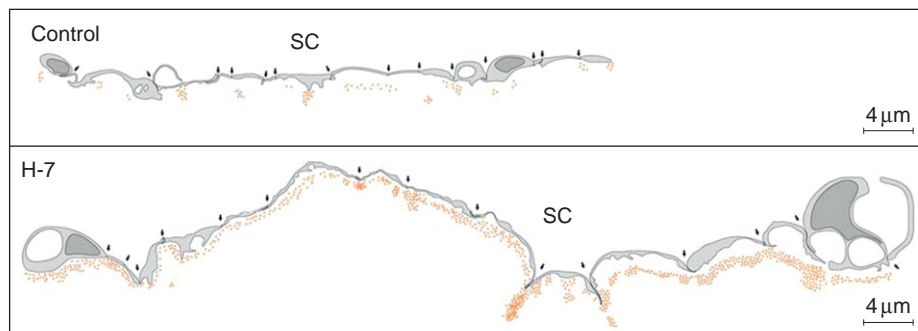


Figure 3 Schematic drawing depicting 15-cell stretches (cell–cell junctions marked by arrows) along Schlemm's canal (SC) and distributions of perfused gold particles that crossed the juxtacanalicular area of control and H-7-treated monkey eyes. Location of individual gold particles represented by red dots. Modified from Sabanay, I., Gabelt, B. T., Tian, B., Kaufman, P. L., and Geiger, B. (2000). H-7 effects on the structure and fluid conductance of monkey trabecular meshwork. *Archives of Ophthalmology* 118: 955–962, with permission from American Medical Association.

Y-27632, the inner wall of Schlemm's canal and the JCT connective tissue are significantly distended compared to control eyes, with discernible separation between the inner wall and JCT connective tissue (Figure 4). The average percent effective filtration length of the inner wall of Schlemm's canal (filtration length/total length $\times 100$) is threefold larger in Y-27632-treated eyes than in controls. A significant positive correlation is found between the average percent effective filtration length of the inner wall and the average percent separation length (separation length/total length $\times 100$, where separation length is the length exhibiting separations between the JCT connective tissue and inner wall), suggesting that the structural correlate to the increase in outflow facility after Y-27632 is physical separation between the JCT connective tissue and the inner wall of Schlemm's canal. The Y-27632-induced TM structural changes are consistent with those induced by latrunculin B or H-7, further indicating that cellular relaxation and drainage surface expansion in the TM are likely the key mechanisms by which

cytoskeletal agents increase outflow facility. Recently, a ROK β /ROCK-I and ROK α /ROCK-II inhibitor, INS117548, was confirmed to induce a dose-dependent decrease in the number of stress fibers, focal adhesion size, and numbers in HeLaJW cells. INS117548 also significantly lowered IOP in living monkeys. Since prolonged, acute exposure of formulated INS117548 produced no observable effects on the ocular surface at nearly threefold of a minimally effective IOP-lowering dose, INS117548 could be a potential efficacious and well-tolerated IOP-lowering agent for glaucoma therapy.

Myosin II inhibitors (blebbistatin)

Blebbistatin is a highly specific inhibitor of myosin II that inhibits both the adenosine triphosphatase (ATPase) and gliding motility activities of myosin II without inhibiting MLCK. It does not affect ATP binding or hydrolysis, but instead binds to the myosin-ADP-Pi complex, interfering with phosphate release, keeping myosin in an actin-detached state and preventing actomyosin interaction.

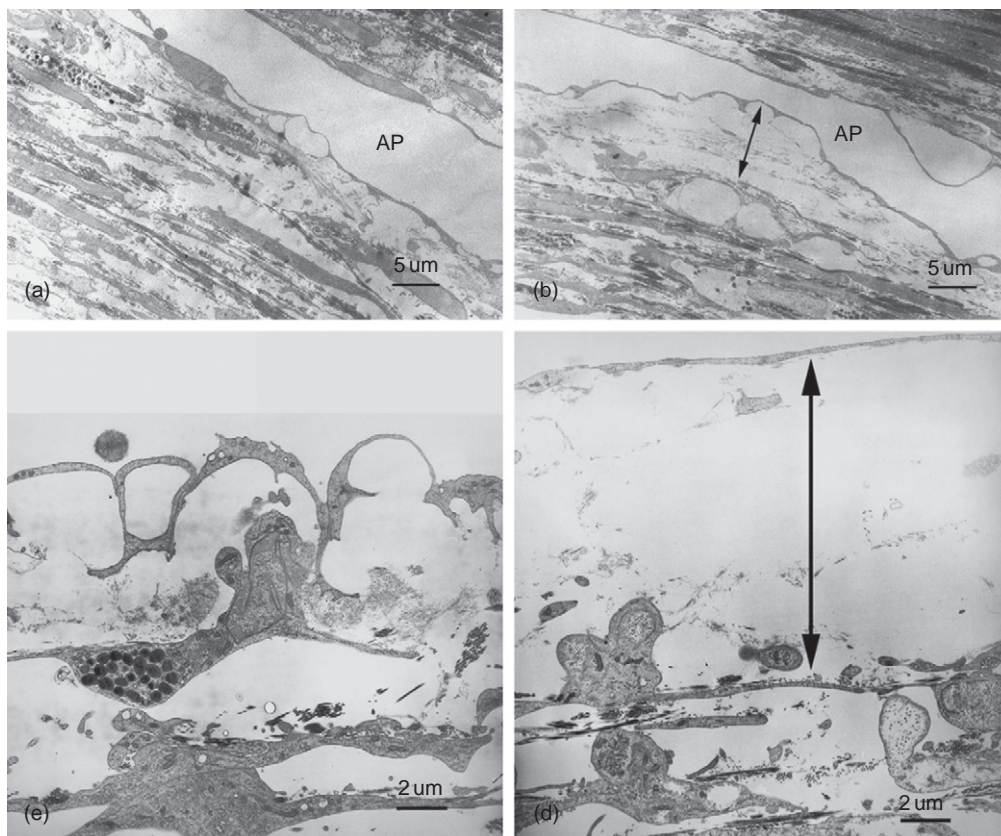


Figure 4 Electron microscopic analysis of the trabecular meshwork of bovine eyes. (a) In a control eye, the inner wall of Schlemm's canal and JCT tissue were more in contact with underlying structures, the JCT tissue appeared compact and without separation between the inner wall and JCT tissue. (b) In a Y-27632-treated-eye, the inner wall appeared distended, the JCT tissue was loose with significant separation between the inner wall and JCT tissue (double arrow). (c) In areas without separation, the connection between the inner wall and JCT cells and between the inner wall and JCT matrix were maintained. (d) In separated areas, the connection between the inner wall and JCT cells and between the inner wall and JCT matrix were lost (double arrow). Modified from Lu, Z., Overby, D. R., Scott, P. A., Fredo, T. F., and Gong, H. (2008). The mechanism of increasing outflow facility by rho-kinase inhibition with Y-27632 in bovine eyes. *Experimental Eye Research* 86: 271–281. Copyright Elsevier.

Confluent cultures of primary porcine TM cells treated with blebbistatin in the presence of serum have revealed dose-dependent changes in cell morphology, decreases in actin stress fibers, and in focal contacts and cell–cell adherens junctions. Perfusion of anterior segments of enucleated porcine eyes with blebbistatin significantly increases outflow facility. The effects of blebbistatin on TM cell morphology and actomyosin cytoskeletal organization are consistent with the effects of the inhibitors of Rho kinase, PKC, and MLCK on TM cells. However, unlike these inhibitors, which mediate their effects by inhibiting myosin II activity through decreased MLC phosphorylation, blebbistatin mediates its effects without affecting the phosphorylation status of MLC in TM cells, confirming its specificity to myosin II ATPase activity. Since the integrity of the inner wall of aqueous plexi in blebbistatin-perfused porcine eyes is intact, and the TM cell morphology after the drug appears to be similar to that noted in vehicle-treated eyes, observable structural deterioration in the TM may not be the main mechanism for the drug-induced increase in outflow facility, but rather the cellular relaxation of the trabecular outflow pathway following blebbistatin may be involved.

Actomyosin-Modulating Gene Therapy

Modulating proteins that negatively regulate actin–myosin interactions can also induce TM relaxation. Caldesmon is such a protein, whose function is the regulation of actomyosin contractility. When caldesmon is overexpressed, actin becomes uncoupled from myosin, which can affect both actomyosin-driven contractility and actin polymerization. In addition, exoenzyme C3 transferase may also affect actin–myosin interactions. Rho GTPases are the preferred intracellular targets of exoenzyme C3 transferase. The latter specifically inhibits Rho-GTP at the beginning of the Rho activation cascade, thereby blocking the whole Rho cascade. Adenovirus-delivered exoenzyme C3 transferase (C3-toxin) complementary DNA (cDNA) and nonmuscle caldesmon cDNA have been successfully expressed in cultured human TM cells. Perfusions in organ-cultured human or monkey eyes following overexpression of these genes have shown significant increases in outflow facility. Specific inhibition of Rho-kinase activity in the TM by dominant-negative Rho expression also increases outflow facility in organ-cultured anterior segments of postmortem human eyes. All these suggest that, similar to pharmacological approaches, gene therapies may also inhibit actomyosin system in the TM and in turn increase trabecular outflow facility through blocking the Rho activation pathway and overexpressing modulating proteins.

Microtubule Inhibition

The microtubule system consists of microtubules and associated proteins. Similar to microfilaments, microtubules

are also highly organized in cells of the trabecular outflow route (**Figure 1**). They are not intrinsically contractile, but are important for directional cell motility and driven by specific microtubule motor proteins for cytoplasmic trafficking of vesicles and organelles. Associated proteins that bind to microtubules can affect the latter's stability and potentially attach the latter to other cytoskeletal filaments (e.g., microfilaments). Microtubule function could affect outflow pathway events through direct cellular mechanical effects (e.g., tensegrity), influences on ECM or cell membrane turnover (through vesicle movement), or through secondary signaling (e.g., leading to activation of the actin cytoskeleton).

Ethacrynic acid, known to be a potent microtubule inhibitor, reduces outflow resistance in enucleated calf and human eyes and in living monkey eyes, and concomitantly reduces IOP in live rabbit, monkey, and human eyes. Although ethacrynic acid primarily inhibits microtubule assembly, it also induces a rapid decrease in phosphotyrosine levels of focal adhesion kinase and a more subtle decrease in paxillin phosphorylation. Dephosphorylation of these proteins disrupts signaling pathways that normally maintain the stability of the actin microfilaments and cellular adhesions, and consequently induces the onset of retraction, stress fiber disruption, or complete disruption of focal adhesions. This indicates a close relationship between the microtubule system and the actomyosin system. Several new derivatives of ethacrynic acid significantly decrease IOP in cats and monkeys. These derivatives are more potent than ethacrynic acid in terms of inducing cell-shape alterations and decreasing actin stress fibers in human TM cells, suggesting that microtubule disruption may reduce outflow resistance at least partially through perturbation of the actomyosin system.

Significance of Actomyosin Inhibition in Glaucoma Therapy

Glaucoma is one of the most common causes of irreversible blindness in the world. At present, the only effective approach available to treat glaucoma is to reduce IOP. Pharmacologically reducing IOP is usually the first choice in glaucoma therapy. Medications used clinically to decrease IOP include aqueous humor secretory inhibitors (e.g., beta-adrenergic receptor antagonists, alpha2-adrenergic agonists, and carbonic anhydrase inhibitors), uveoscleral-outflow enhancers (e.g., prostaglandin analogs), cholinergic drugs that affect trabecular outflow indirectly by contracting the ciliary muscle and deforming the TM, and epinephrine drugs that work on both the TM (inducing changes in cell shape through a beta-adrenergic receptor-cyclic adenosine monophosphate (cAMP)/PKA-mediated cellular relaxation) and the uveoscleral (mediating endogenously synthesized prostaglandins) outflow routes. Secretory

suppression may affect supplies of oxygen and nutrients to the nonvascularized cornea, lens, and TM. Prostaglandin analogs do not substantially improve trabecular outflow. Cholinergic drug effects on the pupil and accommodation limit their clinical use. Epinephrine-like drugs are no longer used clinically because of their local and systemic side effects. Thus, there are no TM-selective outflow enhancers in current clinical use.

Since evidence has shown, as discussed in this article, that cytoskeletal agents or relevant gene therapies decrease outflow resistance by a mechanism directly related to the TM/Schlemm's canal, pharmacological or genetic perturbation of the actomyosin system in the TM may have potential to open a new avenue in glaucoma treatment. However, although cytoskeletal drugs effectively increase outflow facility and decrease IOP, they could, in principle, have detrimental effects on other anterior segment tissues, especially the cornea. Lower drug concentrations in larger volumes could minimize corneal toxicity without significantly sacrificing the drug's effect on the TM following topical administration. However, the potential cornea toxicity is still an obstacle to the use of higher concentrations of the drugs topically for a greater outflow facility increase. To overcome this problem, novel methods of drug delivery need to be developed. Receptors might be different in different cell types or ECM; therefore, a better understanding of the biomolecular differences between cornea and TM, the different molecular targets or mechanisms for different actin-disrupting agents, and a pro-drug, gene therapy or other site-activated approach, could facilitate the development of TM-selective drugs that reduce outflow resistance without affecting other ocular tissues.

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The authors would also like to disclose here that the University of Wisconsin and the Weizmann Institute of Science hold a patent related to cytoskeletal compounds, (latrunculins and H-7), and that patent has been licensed by Inspire Pharmaceuticals; accordingly, Drs. Kaufman (UW) and Geiger (WIS) have a proprietary interest. Additionally, Inspire conducts research in Dr. Geiger's and Dr. Kaufman's laboratories, and both Dr. Kaufman and Dr. Geiger serve as advisors/consultants to Inspire. Inspire also provides unrestricted support for research in Dr. Kaufman's laboratory.

See also: Biological Properties of the Trabecular Meshwork Cells; The Biology of Schlemm's Canal; Biomechanics of Aqueous Humor Outflow Resistance; The Fibrillar Extracellular Matrix of the Trabecular Meshwork; Functional Morphology of the Trabecular Meshwork; Pharmacology of the Aqueous Humor Outflow; Regulation of Extracellular Matrix Turnover in the Aqueous Humor Outflow Pathways; Role of Proteoglycans in the Trabecular Meshwork; Structural Changes in the Trabecular Meshwork with Primary Open Angle Glaucoma.

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