Signaling Mechanism of TGF-β1–Induced Collagen Contraction Mediated by Bovine Trabecular Meshwork Cells

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PURPOSE. To characterize the intracellular signaling mechanism that underlies the contraction of trabecular meshwork (TM) tissue.

METHODS. The contraction of collagen mediated by bovine TM cells was evaluated by measuring changes in the diameter of collagen gels in which the cells were embedded. Changes in the organization of the actin cytoskeleton were examined by laser-scanning confocal microscopy of cells stained with fluorescent phalloidin. Cell motility was monitored by time-lapse video microscopy.

RESULTS. Transforming growth factor (TGF)-β1 induced marked TM-cell–mediated contraction of collagen gels in a concentration- and time-dependent manner. Inhibitors of protein kinase C (PKC) blocked this effect of TGF-β1, whereas an inhibitor of PKA and -G did not. An inhibitor of the small guanosine triphosphatase (GTPase) Rho also inhibited TGF-β1–induced collagen contraction, whereas an activator of Rho promoted this effect of TGF-β1. Furthermore, inhibition either of the release of Ca2+ from internal stores or of the activation of myosin light-chain kinase (MLCK) prevented gel contraction in response to TGF-β1. The effects of these various agents on TGF-β1–induced contraction of collagen gels mediated by TM cells were mirrored by their effects on TGF-β1–induced formation of actin stress fibers, cell spreading (the extension of cellular processes), and cell motility under conditions in which cell contraction was not possible.

CONCLUSIONS. TGF-β1 induces TM-cell–mediated collagen gel contraction through activation of Rho and the Ca2+-dependent enzymes PKC and MLCK. These same signaling molecules contribute to TGF-β1–induced rearrangement of the actin cytoskeleton, cell spreading, and cell motility. (Invest Ophtalmol Vis Sci. 2002;43:3465–3472)

Trabecular meshwork (TM) tissue is located at the angle of the anterior chamber, which is the main pathway by which aqueous humor exits the eye to maintain intraocular pressure. This outflow from the anterior chamber, through the TM tissue, to the episcleral vein is a pressure-dependent filtration process that does not change the solute composition of the aqueous humor. The contraction of TM tissue is thought to be responsible for the outflow facility. TM tissue consists mostly of TM cells and various extracellular matrix (ECM) proteins, including collagens (types I, III, IV, and V), proteoglycans, laminin, fibronectin, and elastin. TM cells appear to perform various biological functions, including phagocytosis, chemotaxis, and ECM protein synthesis, secretion, and degradation. The contraction of TM tissue is probably mediated by the interaction of TM cells with extracellular collagen.

Human skin fibroblasts embedded in gels composed of collagen type I mediate contraction of the collagen fibrils if the latter are not attached to the experimental container. Various growth factors induce the contraction of type I collagen gels in which human keratocytes are cultured in three dimensions. Transforming growth factor (TGF)-β1 induces the contraction of human skin fibroblasts, human Tenon's capsule fibroblasts,12 and bovine keratocytes. TGF-β1 is a member of the TGF-β family of multifunctional growth factors, with receptors that possess a serine-threonine kinase domain.

Contraction and relaxation of TM tissue are thought to control intraocular pressure and are mediated by intracellular signal-transduction systems. Inhibition of protein kinase C (PKC), a serine-threonine kinase, thus induces the relaxation of TM tissue in vitro, and Rho-associated coiled coil-forming protein kinase (ROCK), which is a target of the small guanosine triphosphatase (GTPase) Rho, is implicated in TM contraction in vitro. Moreover, serine-threonine kinase inhibitors increase aqueous humor outflow through TM tissue in vitro, and administration of the ROCK inhibitor Y-27632 to rabbits reduces intraocular pressure and increased outflow facility in vivo. These observations suggest that the PKC and Rho signaling pathways participate in the regulation of conventional outflow facility and intraocular pressure. We investigated the intracellular signaling mechanisms that underlie contraction of TM tissue with the use of three-dimensional cultures of bovine TM cells embedded in type I collagen gels. Our results showed that TGF-β1 induced the contraction of collagen gels mediated by these cells, and we described the effects of various inhibitors and activators of signaling molecules on this process. We also evaluated the effects of these inhibitors and activators on TGF-β1–induced changes in the cytoskeletal organization and motility of TM cells.

METHODS

Materials

Eagle's minimum essential medium (MEM), fetal bovine serum (FBS), and trypsin-EDTA (0.5% trypsin; 5.3 mM EDTA, tetrasodium salt) were obtained from Gibco BRL (Rockville, MD). Acid-solubilized porcine type I collagen (Cellmatrix type I-A; 3 mg/mL) and reconstitution buffer (50 mM NaOH, 260 mM NaHCO3, and 200 mM HEPES [pH 7.3]) were from Nitta Gelatin (Yao, Osaka, Japan). Dulbecco's phosphate-buffered saline (PBS) was from Nissui Pharmaceutical (Tokyo, Japan), and bovine serum albumin (fraction V; BSA) was from Nacalai Tesque (Kyoto, Japan). Paraformaldehyde, recombinant human platelet-de-
rived growth factor-BB (PDGF-BB), recombinant human basic fibroblast growth factor (bFGF), phorbol 12-myristate 13-acetate (PMA), 4α-phorbol 12-myristate 13-acetate (4α-PMA), 1-(5-isoquinolinesulfonyl)-2-methyl-piperazine (H-7), 1,2-dimethoxy-V-methyl(1,3)benzodioxolo[5,6-c]phenanthridinium chloride (chelerythrine), 3-(1-(dimethylaminopropyl)imidazol-1-yl)-4-(1H)maleimide (bisindolylmaleimide I), N-(2-guanidinoethyl)-5-isoquinolinesulfonamide (HA1004), 1α-hydroxyl phosphatidic acid (LPA), 1-(5-chloronaphthalene-1-sulfonyle)-1H-benzo-hydro-1,4-diazepine (ML-9), 1-(N-Obis[5-isoquinolinesulfonyl]N-vmethyly-1-tirosyl)-4-phenylpiperazine (KN-62), and cytochalasin D were obtained from Sigma (St. Louis, MO). Exocytome C3 was from Upstate Biotechnology (Lake Placid, NY). Recombinant human TGF-β1 was from R&D Systems (Minneapolis, MN), and recombinant human insulin-like growth factor (IGF)-I and recombinant human epidermal growth factor (EGF) were from Collaborative Biomedical Products (Bedford, MA). Twenty-four-well cell culture clusters and 162-cm² cell culture flasks were obtained from Corning (Corning, NY), and glass-bottomed dishes were from Matsunami (Kishiwada, Osaka, Japan). Thapsigargin and 2-aminooxothiophenyloborates (2-APB) were obtained from Calbiochem-Novabiochem (Darmstadt, Germany), and fluorescent phalloidin (Alexa Fluor 568) was from Molecular Probes (Eugene, OR).

Isolation of Bovine TM Cells

TM cells were isolated as described20,21 from fresh bovine eyes obtained from a local abattoir. They were cultured under 5% CO₂ in MEM, reconstitution buffer, TM cell suspension, and water were mixed in an ice bath at a ratio of 7:1:1:1:1 (see Materials and Methods for full details). Bovine TM cells were collected by treatment of cultures with trypsin-EDTA for 5 minutes, washed twice with unsupplemented MEM, and resuspended in MEM supplemented with 10% FBS. The experiments were performed with cells subjected to five passages. Three different TM cell lines were established.

Contraction Assay

The collagen gel contraction assay was performed as described previously,7,22–25 with some modifications. The wells of 24-well cell culture clusters were each coated with 1 ml 1% BSA for 1 hour at 37°C. Bovine TM cells were collected by treatment of cultures with trypsin-EDTA for 5 minutes, washed twice with unsupplemented MEM, and resuspended in MEM at a density of 2.2 × 10⁶ cells/mL. The resultant mixture (0.5 mL) was added to each well of the BSA-coated cell culture clusters, and formation of a collagen gel was induced by incubation at 37°C under 5% CO₂ for 90 minutes. MEM (0.5 mL), with or without various agents, was then added on top of the collagen gels, and, after 1 hour, the gels were freed from the walls of the culture wells with the use of a micropatula. The diameter of the collagen gel was measured with a ruler every 24 hours for 5 or 6 days. The extent of contraction of the collagen gels mediated by the TM cells was expressed in terms of the decrease in gel diameter compared with the initial diameter. Each experiment was performed in triplicate and was repeated at least twice with different TM cell lines.

Phalloidin Staining

TM cells were cultured in collagen gels (2 × 10⁶ cells/mL) and glass-bottomed dishes (not coated with BSA). The collagen solution containing TM cells was prepared as described for the contraction assay, and a 50-μL portion was spread on the bottom of the dish. After formation, the collagen gel was overlaid with 2 ml MEM in the absence or presence of TGF-β1 (1 ng/mL) and test agents. The cells were cultured for 2 days, washed with PBS, and fixed for 30 minutes at room temperature with 1% paraformaldehyde in PBS. Fixed cells were washed with PBS, air-dried, and incubated for 30 minutes at room temperature with PBS containing 1% BSA to block nonspecific binding sites. After washing with PBS, the cells were incubated with fluorescent phalloidin (5 U/mL in PBS containing 1% BSA; Alexa Fluor 568; Molecular Probes) for 30 minutes at room temperature, washed with PBS containing 0.1% Tween 20, and observed with a laser-scanning confocal microscope (Fluoview; Olympus, Tokyo, Japan).

Time-Lapse Video Microscopy

The motility of bovine TM cells was monitored by time-lapse video microscopy (Axiovision system with CO₂ incubator; Carl Zeiss, Jena, Germany). TM cells were cultured in collagen gels and glass-bottomed dishes. The collagen solution containing the cells was prepared as has been described, with the exception that the cell density was 4 × 10⁴ cells/mL, and 50 μL of the solution was spread on the bottom of the dish. After formation of the collagen gel, it was overlaid with 4 mL MEM containing 1% FBS in the absence or presence of TGF-β1 (1 ng/mL) and test agents. After culture for 24 hours, the cells were examined by time-lapse video microscopy for an additional 12 hours. Cell behavior was evaluated by one of the authors who was blinded to the specific culture conditions.

Statistical Analysis

Data were analyzed by the Dunnett multiple comparison test. P < 0.05 was considered statistically significant.

RESULTS

Effects of Growth Factors on Collagen Contraction Mediated by Bovine TM Cells

We first examined the effect of TGF-β1 on collagen contraction mediated by bovine TM cells cultured in a three-dimensional collagen gel by measuring the change in diameter of the gel. TGF-β1 induced TM-cell-mediated collagen contraction in a time- and concentration-dependent manner (Figs. 1, 2A). The effects of other growth factors on TM-cell-mediated collagen contraction were also examined. PDGF-BB induced a small but significant contraction at the highest concentration (10 ng/mL) examined, whereas EGF, IGF-I, and bFGF had no effect (Figs. 2B–E). To verify that the TGF-β1-induced decrease in collagen gel diameter did not reflect collagen degradation, we examined the extent of collagen breakdown by measuring the amount of hydroxyproline produced after acid-heat hydrolysis of the medium; culture of cells with TGF-β1 (0.01–10 ng/mL) had no effect on the amount of hydroxyproline detected (data not shown). The reduction in size of the collagen gel induced by TGF-β1 thus reflected contraction of the gel and not its degradation.

Role of Protein Kinases in TGF-β1–Induced Collagen Contraction Mediated by TM Cells

To investigate which signal transduction systems participate in the TGF-β1–induced contraction of collagen gels mediated by bovine TM cells, we examined the effects of various inhibitors of protein kinases in the presence of TGF-β1 at 1 ng/mL. The PKC inhibitor bisindolylmaleimide I (BIM) reduced the extent of TGF-β1-induced collagen contraction in a dose-dependent manner, with the inhibitory effect being statistically significant at concentrations of 1 and 10 μM (Fig. 3A). The PKC inhibitors H-7 and chelerythrine also each exhibited similar dose-dependent inhibitory effects on TGF-β1–induced collagen contraction (data not shown). In contrast, HA1004, an inhibitor of protein kinases A (PKA) and G (PKG), reduced the extent of TGF-β1-induced collagen contraction at concentrations of 0.1 to 100 μM (Fig. 3B). These results thus suggest that PKC, but not PKA or PKG, contributes to TM-cell-mediated collagen contraction triggered by TGF-β1.

We next determined whether the activation of PKC was sufficient to induce TM-cell–mediated collagen gel contraction. Neither the PKC activator PMA nor its inactive analogue 4a-PMA, at concentrations of 0.1 to 100 nM, affected collagen...
FIGURE 1. Effect of TGF-β1 on collagen gel contraction mediated by bovine TM cells. (A) Time-course and dose dependence of TGF-β1-induced collagen gel contraction. Cells were cultured in collagen gels for the indicated times in the absence (○) or presence of TGF-β1 at concentrations of 0.01 ng/ml (●), 0.1 ng/ml (▲), 1 ng/ml (▲), or 10 ng/ml (▲). The change in diameter of the collagen gels during culture was measured. Data are means ± SD of triplicate results of a representative experiment. (B) Representative photographs of collagen gel cultures of TM cells incubated for 6 days with the indicated concentrations of TGF-β1.

contraction in the absence or presence of TGF-β1 (Fig. 4). Thus, although PKC appears to participate in TM-cell-mediated contraction of collagen gels induced by TGF-β1, the activation of this enzyme by itself does not elicit such contraction.

Given that PKC and various other protein kinases are dependent on Ca²⁺ for their activation, we examined the possible role of Ca²⁺ in TGF-β1-induced collagen contraction mediated by bovine TM cells. Thapsigargin and 2-APB, both of which block the release of Ca²⁺ from the endoplasmic reticulum, each inhibited the stimulatory effect of TGF-β1 on collagen contraction in a concentration-dependent manner (Fig. 5); neither agent affected collagen gel contraction in the absence of TGF-β1. The possible role of other Ca²⁺-dependent protein kinases was also investigated. Whereas KN-62, an inhibitor of Ca²⁺-and calmodulin-dependent protein kinase II (CaM kinase II), had no effect on collagen contraction in the absence or presence of TGF-β1 (Fig. 6A), the myosin light-chain kinase (MLCK) inhibitor ML-9 reduced the extent of TGF-β1-induced collagen contraction in a concentration-dependent manner without affecting the basal level of contraction (Fig. 6B). These results thus suggest that the release of Ca²⁺ from the endoplasmic reticulum is required for TGF-β1-induced collagen contraction mediated by bovine TM cells, and that, in addition to PKC, MLCK (but not CaM kinase II) contributes to this process.

Role of Rho and Actin Polymerization in TGF-β1-Induced Collagen Contraction Mediated by TM Cells

We next examined the potential contribution of Rho to TGF-β1–induced collagen contraction mediated by bovine TM cells. The Rho activator LPA potentiated the stimulatory effect of TGF-β1 on collagen contraction in a concentration-dependent manner (Fig. 7A). This action of LPA was significant at concentrations of 1 and 10 μM. Conversely, exoenzyme C3, an inhibitor of Rho, reduced the extent of collagen contraction induced by TGF-β1, also in a concentration-dependent manner (Fig. 7B). This effect of exoenzyme C3 was significant, although relatively small, at concentrations of 1 and 2 μg/mL.

Neither LPA nor exoenzyme C3 affected TM-cell-mediated collagen contraction in the absence of TGF-β1. These results thus suggest that Rho contributes to TGF-β1-induced collagen contraction mediated by TM cells, but that the activation of this GTPase is not sufficient by itself to trigger contraction.

Given that Rho plays an important role in rearrangement of the actin cytoskeleton, we investigated the effect of cytochalasin D, an inhibitor of actin polymerization, on TM-cell-mediated collagen contraction. Cytochalasin D inhibited in a concentration-dependent manner the TM-cell-mediated collagen contraction elicited by TGF-β1, although it had no effect on contraction of collagen in the absence of TGF-β1 (Fig. 8). These results thus suggest that reorganization of the actin cytoskeleton plays an important role in TGF-β1-induced collagen contraction mediated by bovine TM cells.

Regulation of Cytoskeletal Reorganization in Bovine TM Cells

Given that our results implicated reorganization of the actin cytoskeleton in TM-cell-mediated collagen contraction induced by TGF-β1, we next examined changes in the cytoskeleton by staining of actin with fluorescent phalloidin (Alexa Fluor 568; Molecular Probes) and laser-scanning confocal microscopy after culture of cells for 2 days in collagen gels. These experiments were performed with three-dimensional cultures in dishes not coated with BSA to prevent collagen gel contraction. In cells cultured in collagen gels in the absence of supplements, actin filaments were observed as dots at the cell surface and in the cytosol (Figs. 9A, 9B). In contrast, the presence of TGF-β1 induced both cell spreading and the formation of thick stress fibers (Figs. 9C, 9D). Although the PKC activator PMA did not affect the pattern of actin filaments observed in the presence of TGF-β1 (Figs. 9E, 9F), the PKC inhibitor BIM reduced the extents of both cell spreading and formation of stress fibers induced by TGF-β1 (Figs. 9G, 9H). The Rho activator LPA potentiated the effects of TGF-β1 on both cell spreading and formation of stress fibers (Figs. 9I, 9J), whereas the Rho inhibitor exoenzyme C3 blocked these effects of TGF-β1 (Figs. 9K, 9L). Inhibition of actin polymerization by
cytochalasin D in TGF-β1-treated cells resulted in marked disruption of the cytoskeletal system. No stress fibers were detected, but dense aggregates of actin were apparent (Figs. 9M, 9N). Overall, these results thus suggest that PKC and Rho contribute to the reorganization of the actin cytoskeleton induced by TGF-β1 in bovine TM cells.

Time-Lapse Video Microscopy of Bovine TM Cell Motility

Finally, given that reorganization of the actin cytoskeleton contributes to cell motility, we examined the motility of bovine TM cells by time-lapse video microscopy. Cells were cultured in collagen gels in the presence of 1% FBS. Again, the culture dishes were not coated with BSA to prevent contraction of the collagen gel. In the presence of FBS alone, the cell somas moved in three dimensions within the collagen gel. The cells expanded and extended several cellular processes, at the tips of which several finer extensions were observed (Fig. 10A). These processes were subsequently retracted and replaced by others. By the addition of TGF-β1, the movement of the cell somas and the protrusion of cellular processes with finer extensions at their tips was also observed, although the soma of cells cultured with this growth factor was larger than that of those cultured without it (Fig. 10B). These results suggest that TGF-β1 does not affect the overall movement of the cells and the active protrusion and retraction of cellular processes. The further addition of the PKC activator PMA had no effect on cell movement (Fig. 10C). In contrast, in the presence of TGF-β1, the PKC inhibitor BIM induced cell rounding and immobilization, although the cellular processes and finer extensions were still apparent (Fig. 10D). In the presence of TGF-β1, the Rho

![Figure 2](https://example.com/figure2.png)

**Figure 2.** Effects of various growth factors on collagen gel contraction mediated by bovine TM cells. Cells were cultured in collagen gels for 5 days in the presence of the indicated concentrations of (A) TGF-β1, (B) EGF, (C) IGF-I, (D) PDGF-BB, or (E) bFGF, after which the change in diameter of the collagen gels was measured. Data are the mean ± SD of triplicate results of representative experiments. *P < 0.001 versus absence of growth factor.

![Figure 3](https://example.com/figure3.png)

**Figure 3.** Effects of the PKC inhibitor BIM (A) and the PKA and PKG inhibitor HA1004 (B) on TGF-β1-induced collagen gel contraction mediated by bovine TM cells. Cells were cultured in collagen gels in the presence of TGF-β1 (1 ng/mL) and the indicated concentrations of BIM or HA1004 (●). Cells were also cultured in the absence of TGF-β1, BIM, and HA1004 (○). The change in collagen gel diameter was assessed after 5 days. Data are the mean ± SD of triplicate results of representative experiments. *P < 0.001 versus TGF-β1 alone.

![Figure 4](https://example.com/figure4.png)

**Figure 4.** Effects of the PKC activator PMA and its inactive analogue 4α-PMA on collagen gel contraction mediated by bovine TM cells. Cells were cultured in collagen gels for 5 days in the absence (open symbols) or presence (closed symbols) of TGF-β1 (1 ng/mL) as well as in the presence of the indicated concentrations of PMA (circles) or 4α-PMA (triangles), after which the change in collagen gel diameter was measured. Data are the mean ± SD of triplicate results of a representative experiment.
The effect of TGF-β1 and bFGF had a substantial effect on TM-cell determinant of cell shape and motility. EGF, IGF-I, PDGF-BB, and bFGF increased the abundance of TGF-β1 mRNA in porcine TM cells. TGF-β1 may promote the contraction of TM cells in vivo. We therefore suggest that TGF-β1 may contribute to the regulation of intraocular pressure through such an effect. TGF-β1 also stimulates collagen contraction mediated by other cell types, including corneal fibroblasts, skin fibroblasts, and mouse embryonic fibroblasts. However, not only TGF-β1 but also EGF, IGF-I, PDGF-BB, and bFGF stimulate collagen contraction mediated by corneal fibroblasts. In addition to TGF-β1, IGF-I stimulates collagen contraction mediated by skin fibroblasts, and EGF antagonizes TGF-β1–induced collagen contraction mediated by these cells. Tissue contraction may thus be regulated differently in the cornea, skin, and TM.

The physiological significance of our observation that TGF-β1 stimulates the contraction of collagen gels mediated by cultured bovine TM cells remains to be determined. However, the concentration of TGF-β2 in aqueous humor has been shown to be increased in patients with glaucomatous eyes. Given that TGF-β2 increases the abundance of TGF-β1 mRNA in porcine TM cells, TGF-β1 may promote the contraction of TM cells in vivo. We therefore suggest that TGF-β1 may contribute to the regulation of intraocular pressure through such an effect. TGF-β1 also stimulates collagen contraction mediated by other cell types, including corneal fibroblasts, skin fibroblasts, and mouse embryonic fibroblasts. However, not only TGF-β1 but also EGF, IGF-I, PDGF-BB, and bFGF stimulate collagen contraction mediated by corneal fibroblasts. In addition to TGF-β1, IGF-I stimulates collagen contraction mediated by skin fibroblasts, and EGF antagonizes TGF-β1–induced collagen contraction mediated by these cells. Tissue contraction may thus be regulated differently in the cornea, skin, and TM.
that PKC is an important mediator of TGF-β1 action.\textsuperscript{33} Overall, our results suggest that the contraction of collagen gels mediated by bovine TM cells in response to TGF-β1 is mediated by Ca\textsuperscript{2+}, the Ca\textsuperscript{2+}-dependent enzymes PKC and MLCK, and Rho. TGF-β1 also induces Rho activation in mammary epithelial cells.\textsuperscript{34} The contraction of smooth muscle cells is promoted by phosphorylation of myosin light chain, which is mediated by MLCK. The phosphorylation level of myosin light chain is also increased as a result of inhibition of myosin light-chain phosphatase, and this enzyme is inhibited by Rho activation\textsuperscript{35} and by PKC activation,\textsuperscript{36} both of which are now implicated in TGF-β1–induced collagen contraction mediated by TM cells.

Rho is an important regulator of the actin cytoskeleton.\textsuperscript{37,38} TGF-β1 has previously been shown to increase the expression of α-smooth muscle actin in TM cells.\textsuperscript{39} Consistent with the results of our gel contraction experiments, laser-scanning confocal microscopy of TM cells stained with fluorescent phalloidin revealed that TGF-β1 induced cell spreading and the formation of stress fibers in cells cultured in collagen gels under conditions not permissive for contraction of extracellular collagen, and that these effects of TGF-β1 were promoted by LPA and inhibited by BIM, exoenzyme C3, or cytochalasin D.

Time-lapse video microscopy of bovine TM cells cultured in collagen gels revealed that these changes in the actin cytoskeleton were also related to cell motility. Thus, in the presence of TGF-β1, the cell somas moved in three dimensions and extended many processes. Again, the movement of the cell somas and the extension of cellular processes were promoted by LPA.
and inhibited by BIM, exoenzyme C3, or cytochalasin D. Our results therefore suggest that the movement of TM cell somas in collagen gels is mediated by the formation of stress fibers and is closely related to the contraction observed when the gels are not attached to the walls of the culture vessel. Previous studies have shown that inhibitors of PKC and of ROCK induce relaxation of strips of TM tissue. Furthermore, MLCK has been implicated in aqueous humor outflow through the TM. Our collagen gel contraction assay with isolated TM cells allows the effects of different agents (at various concentrations) on cell contraction to be investigated much more readily compared with the systems used for these previous studies. With this assay, we have now provided new insight into the intracellular signaling mechanisms that underlie the changes in the interaction of TM cells with the ECM triggered by TGF-β1 and which may contribute to the regulation of aqueous humor outflow in vivo.

The contraction and relaxation of the TM appear to regulate intraocular pressure by changing the space at the angle available for the outflow of aqueous humor. Intraocular pressure has been shown to be reduced and outflow facility to be increased as a result of TM relaxation induced by inhibitors of PKC, ROCK, or cytoskeletal reorganization. Our present data showing that TGF-β1 promotes TM-cell–mediated collagen contraction through both activation of signaling by PKC and Rho and reorganization of the cytoskeleton are in agreement with these previous observations.

References


