Review

Ocular Outflow Facility with Emphasis on Neuronal Regulation of Intraocular Smooth Muscles

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The narrow range of intraocular pressure (IOP) (12–20 mmHg) in normal individuals has stimulated a search for possible regulatory mechanisms^{7,18,65)} of aqueous production and outflow^{6,46)}. Compared with aqueous production, the aqueous outflow mechanisms of the neuronal, humoral, and mechanical processes have been studied much less. Because the eye constitutes a small portion of total body mass, it is very difficult to determine the neuronal and mechanical regulations of intraocular muscle and outflow facility. Locally acting mechanisms, should be an ideal means for integrating its physiology^{58,76)}. The peripheral nervous system is designed to effect such local control. Because most available antiglaucoma agents interact with the autonomic mechanisms and mechanical activities of the smooth muscles in the eye^{53,59,68)}, combined studies of the intraocular muscles with eye perfusion^{7,66)} and cell shape changes of cultured cells from the outflow route^{22,66)} would suggest the role of the nervous system in regulating IOP.

From an historical view point, much speculation and minimal experimentation have been focused on the influence of the iris sphincter, the iris dilator, and the ciliary muscles on aqueous humor outflow. Accommodation, cholinergic agonists, and stimulation of the oculomotor nerve, all increase outflow facility¹⁹⁾, whereas ganglionic blocking agents and anticholinergic drugs decrease the ocular outflow facility^{5,66)}. Furthermore, the outflow facility increase with intravenous pilocarpine administration is instantaneous, suggesting that the effect is mediated by an arterially perfused tissue^{31,32,46)}. Following disinsertion of the ciliary muscle, the outflow facility did not increase with the application of pilocarpine³²⁾. All things considered, these findings suggest that intraocular muscle tone somehow alters the meshwork so as to increase outflow facility during ciliary muscle contraction and decrease facility during ciliary muscle relaxation⁶⁶⁾.

Because the primary function of the trabecular meshwork is to maintain the appropriate aqueous humor outflow rate and IOP, the focus of outflow pathway research should be on the systems that modulate aqueous outflow. Barany⁴⁾, Grant^{25,26)}, Bill^{5,6)}, Epstein^{17,18)} and Kaufman^{31,32)} greatly contributed to this body of knowledge.

The outflow resistance in the conventional outflow route can be altered by changes in intraocular muscle tone^{35,59,72)} and direct effects on the trabecular meshwork^{48,66,70)}. In the present review, neuronal and mechanical influences on the intraocular muscle and outflow facility of the aqueous humor are discussed.

Open-angle glaucoma or angle-closure glaucoma ?

The iris sphincter and ciliary muscles are primarily cholinergically innervated and contract in response to muscarinic agonists. In primates, the iris inserts into the ciliary muscle posterior to the scleral spur, and the ciliary muscle inserts at the scleral spur and at the posterior inner aspect of the trabecular meshwork^{39,69}.

When iridectomy or an opening in the peripheral iris is performed on an eye with angle-closure glaucoma, the new opening in the iris allows the iris to fall away from the angle, giving free access to the trabecular area and avoiding pupillary block formation (Fig. 1). Thus, because iridectomy theoretically relieves most relative pupillary block and closure of the anterior chamber angle, in the present review, only open-angle glaucoma is discussed as glaucoma.

Why does IOP increase ?

As has been well described¹⁸, Table 1 illustrates the relationship between IOP, the coefficient rate of outflow facility (C)^{20,29}, and episcleral venous pressure (Pe). The experimental method to determine the outflow facility of the eye is illustrated in Figs. 2 and 3. An advanced method to determine the outflow facility was developed (Fig. 2)⁶⁵. However, outflow facility

Fig. 1. Primary angle-closure glaucoma (PACG) and primary open-angle glaucoma (POAG). Bulged iris or pupillary block glaucoma is relieved by iridectomy, thus PACG theoretically changes to open-angle glaucoma. Top, PACG; bottom, POAG.

Fig. 2. Ocular perfusion method. An advanced type⁶⁵⁾. The eye can be perfused with various pressures (Suzuki *et al.*; 1992, 1996).

Fig. 3. Ocular perfusion method. A much simpler type⁶⁵⁾. The eye is perfused with a constant pressure (Suzuki *et al.*; 1992, 1996).

can be measured^{65,66)} using a much simpler method (Fig. 3)⁶⁶⁾. With normal parameter (Table 1), a two-fold change in the rate of aqueous formation causes only a 4 mmHg change in IOP (13 to 17 mmHg). In contrast, in a glaucomatous eye, a two-fold change in the rate of aqueous formation causes a 20 mmHg change in IOP instead of the 4 mmHg change seen in the normal eye. Furthermore, in the glaucoma, an increase in the outflow facility of 0.05 $\mu\text{l}/\text{min}/\text{mmHg}$ reduces the IOP by 15 mmHg (39 to 24 mmHg) in contrast with a 1 mmHg reduction in the normal eye. Thus, glaucomatous eyes have greater pressure variations than normal eyes in response to changes, especially in the outflow facility. Drugs that influence the outflow facility cause greater changes in pressure in glaucomatous than in normal eyes.

The relationship between the extent of closure of the filtration angle by synechia or by apposition of the iris to the trabecular meshwork also is seen in Table 1. Obstruction of half the circumference reduces the outflow facility by 50%, and obstruction of three quarters of the circumference reduces the outflow facility by 75%. Closing the filtration angle by half of the circumference raises the pressure in both eyes, to only 19 mmHg in the normal eye, but to 29 mmHg in the poor normal eye. Similarly, with closure of three quarters of the angle, the difference between the eyes is still more drastic, ie, 29 mmHg compared with 49 mmHg.

Table 1. Examples of difference in the degree to which the IOP is calculated to be affected by changes in flow (F) and facility of outflow (C) in different types of eyes. Originally presented by Chandler and Grant (Epstein) (1994) and modified by Suzuki (1996).

	$\mu\text{l}/\text{min}$ (F)	$\mu\text{l}/\text{min}/\text{mmHg}$ (C)	mmHg (Pe)	mmHg (IOP)
	$(F \div C) + P_e = \text{IOP}$			
Normaal	1.5	0.25	9	15
	1 to 2	0.25	9	13 to 17
	1.5	0.3	9	14
Glaucoma	1.5	0.05	9	39
	1 to 2	0.05	9	29 to 49
	1.5	0.10	9	24
Good normal	1.5	0.3	9	14
1/2 angle closed	1.5	0.15	9	19
3/4 angle closed	1.5	0.075	9	29
Poor normal	1.5	0.15	9	19
1/2 angle closed	1.5	0.075	9	29
3/4 angle closed	1.5	0.0375	9	49
Standard	1.5	0.3	9	14
Inflow	0.75	0.3	9	11.5
	0.375	0.3	9	10.3
	3	0.3	9	19
Outflow	1.5	0.15	9	19
	1.5	0.075	9	29
	1.5	0.038	9	49
	1.5	0.019	9	89

Even if the inflow is one-half or one-quarter, the pressure reduction is only 2.5 mmHg (14 to 11.5 mmHg) or 3.7 mmHg (14 to 10.3 mmHg) (Table 1). If the inflow is doubled, the IOP increase is only 5 mmHg. By contrast, when the outflow facility is decreased from 0.3 to 0.15, 0.075, 0.038, and 0.019, the IOP increase is much greater, ie, 5, 15, 35, and 75 mmHg (14 to 89 mmHg). Therefore, the outflow facility is most important for understanding and curing glaucoma. This is the focus of this review. Nevertheless, experimental studies in this important field are scarce^{7,23,65}.

Trabecular meshwork

The aqueous outflow pathway has not been well studied physiologically, because it is small and difficult to access. According to Acott², the human trabecular meshwork is small (100–150 μg) with few cells (200,000–300,000 per eye) that are highly differentiated and moderately difficult to culture^{11,27,33,66,70}. Human trabecular cells grow poorly from donors over 20 years of age. Human, monkey, and bovine trabecular meshwork extracts, cell cultures, and human corneoscleral organ cultures have been the primary focus of molecular analysis of the outflow pathway^{1,48,66}.

According to Grant²⁶, removal of the trabecular meshwork in enucleated human eyes

Table 2. Intraocular distribution of neurotransmitters and autacoids in mammalian eyes

Neurotransmitters Autacoids	Intraocular muscle	On and around Trabecular meshwork	Drainage route
acetylcholine	+	+	+
BNP*	?	+	—?
CGRP*	+	+	+
cholecystokini	+?	+	?
dopamine	—?	?	+
dynorphin	+?	?	+
endothelin	+	+	?
enkepharin (Leu-, Met-)	+	?	?
galanin	?	+	+
neuropeptide Y	+	+	+
neurotensin	+?	—	+
nitric oxide	+	+	+
norepinephrine	+	—?	+?
PHI*	—?	—	+
prostaglandins	+	+	+
serotonin	+	?	+
substance P	+	+?	+
VIP*	+	+	+

Modified from Stone RA⁶³. The galanin data apply only to the cat, since there is marked species variability. The data on BNP are from the pig eye. Dynorphine was identified in the guinea pig iris.

*CGRP, calcitonin gene-related peptide; BNP, brain natriuretic peptide; PHI, peptide histidine isoleucine.

decreases the outflow resistance by 75%, which indicates that the primary resistance is in the trabecular meshwork, most probably within the cribriform layer. In the monkey eye, the primary pressure decrease occurs between the anterior chamber and Schlemm's canal, whereas there is only a minimal difference in pressure between the canal and the episcleral veins^{5,47}. Most of the resistance is between the anterior chamber and Schlemm's canal²⁵.

Peripheral nerves and IOP

The trabecular meshwork is innervated by both the cholinergic and the adrenergic nervous systems, in addition to sensory innervation⁵⁸. Many different peptidergic (VIP^{57,58}, neuropeptide Y, CGRP²⁸, substance P^{62,80} etc) nerve fibers are present, but the significance of the trabecular innervation in relation to aqueous humor outflow is still largely unknown (Table 2). For example, presumed serotonergic nerve fibers are present in the ciliary body, serotonin is in human aqueous humor, and serotonergic agents alter IOP^{58,71}. The serotonergic antagonist, ketanserin, lowers IOP in humans. Because serotonin is localized in the anterior segment nerves⁵⁸, neuronal or other mechanisms may underlie these pharmacologic effects. The effects on intraocular muscle tissue should be studied.

Leu-enkepharin is another example and probably derives from cholinergic neurons of the ciliary ganglion. Opioid mechanisms^{15,30} appear to influence the IOP, by affecting outflow^{18,26}. As shown in Table 2, there are many peptides and purines distributed in intraocular muscles or the drainage, but most studies have been restricted only to morphology or immunohistochemical staining^{56,58}.

Cholinergic regulation

The roles of the iris and the ciliary muscle in mediating outflow facility²⁰ were clarified by two studies. First, removal of the iris alone did not affect IOP, resting outflow facility, and facility responses to pilocarpine. Therefore, it is clear that the ability of the outflow facility to increase the effects of pilocarpine is not mediated by the iris. Second, after disinsertion of the ciliary muscle from its proximal attachment, pilocarpine did not effect outflow facility. Therefore, several studies concluded that the facility-increasing effects of pilocarpine and probably other cholinergic drugs are the results of ciliary muscle contraction, with no measurable effect directly on the meshwork^{31,32,47,66}. As pilocarpine⁵⁵ increases the outflow facility, it stops the uveoscleral outflow almost completely, whereas atropine increases uveoscleral outflow^{46,47}.

Surgery performed on monkeys is much more difficult than on human eyes. The author is surprised to see the monkey results³¹.

Ciliary muscle and aqueous humor outflow

The attachment of the trabecular meshwork to the anterior parts of the ciliary muscle bundles makes it possible for the ciliary muscle to alter the configuration of the trabecular

Fig. 4. Ciliary muscle contraction. The contraction decreases uveoscleral outflow and increases trabecular outflow (Modified from Nilsson⁴⁷) and originally proposed by Bill⁶).

meshwork⁴⁷. When the ciliary muscle contracts (Fig. 4), the anterior part of the muscle thickens, and the meshwork moves toward the anterior chamber, making the meshwork packing looser⁵. The widening of the spaces between the sheets in the meshwork increases the facility in the meshwork. Simultaneously, the spaces between the ciliary muscle bundles narrow and prevent uveoscleral outflow⁶. In other words, when the ciliary muscle contracts^{36,37,59,78}, the spaces between the muscle bundles obstruct aqueous humor drainage via the uveoscleral route^{6,69}. The effect of trabecular drainage is greater than the effect of the obstruction of uveoscleral drainage, so that the net effect is decreased IOP. On the other hand, when the ciliary muscle relaxes (Fig. 5), the muscle bundle becomes thinner and the spaces

Fig. 5. Ciliary muscle relaxation. The relaxation increases uveoscleral outflow and decreases trabecular outflow (Modified from Nilsson⁴⁷) and originally proposed by Bill⁶).

widen; thus, uveoscleral outflow increases⁴⁷⁾. Therefore, depending on the degree of contraction in the ciliary muscle, the aqueous humor can be redistributed via the conventional or the uveoscleral outflow route (Figs. 4, 5).

The other major function of the ciliary muscle is accommodation. The accommodative myopia induced by cholinomimetic antiglaucoma drugs is a major disadvantage to their use, especially in younger patients. The ciliary muscles show a ripple formation⁵⁹⁾. Recording of the ciliary muscle was first accomplished in bovine tissue. This contractile activity makes the ciliary muscle capable of graded, sustained contractions. Because of the anatomic association between the anterior muscle tendons and the trabecular meshwork, this contraction mechanically deforms the meshwork and widens the canal in such a way that the outflow facility increases^{6,52)}.

Lack of obstruction in the uveoscleral route

Muscarinic agonists do not appear to have any direct facility-increasing effect on the trabecular meshwork. In the author's experiment, carbachol or pilocarpine was administered to bovine eyes, and the outflow facility did not increase. This is very curious, because carbachol and/or pilocarpine contracts human ciliary muscle, thus opening the trabecular meshwork pores. To our great surprise, no *in vitro* experiment has successfully demonstrated that muscarinic agonists increase trabecular outflow.

VIP increases Ach release⁶²⁾ from cholinergic nerve terminals of the bovine ciliary muscle without changing the muscle tone⁵⁹⁾. VIP^{56,71)} relating to the cholinergic nerves, causes a slight increase in outflow facility in the monkey^{45,47)}. Because VIP also is a potent stimulator of the adenylate cyclase system, VIP may increase the facility via cAMP.

Why can aqueous pass through the uveoscleral route? According to Nilsson and Bill⁴⁷⁾, the lack of an epithelial barrier between the anterior chamber and the ciliary body makes it possible for aqueous humor to enter the ciliary body from the chamber angle. The aqueous can pass between the muscle bundles and thus enter the supraciliary and suprachoidal spaces⁶⁾. Outside the eye, the fluid becomes part of the orbital tissue and is partly reabsorbed into the orbital blood vessels.

The uveoscleral outflow is relatively pressure insensitive within the normal range of IOP, because the driving force of uveoscleral outflow is the difference in pressure between the anterior chamber and the suprachoroidal space⁴⁷⁾. The pressure in the suprachoroidal space is slightly lower than the IOP in the monkey. An increase in the IOP causes an almost equal change in the suprachoroidal pressure, thus keeping the uveoscleral outflow pressure relatively constant¹⁶⁾. When the IOP increases, the ciliary muscle and supraciliary space compress, causing decreased outflow facility of the uveoscleral flow^{6,16)}.

Dissociation between outflow facility and accommodation

Longitudinal fibers of the ciliary muscle primarily work on outflow facility, whereas circular fibers subserve accommodation. However, a dissociation was observed between the

effects of pilocarpine on accommodation and outflow facility^{19,20}. Further, even though a normal accommodative response to pilocarpine and physostigmine maintained in the re-innervated monkey eye after ciliary ganglionectomy, outflow facility did not increase in response to pilocarpine^{31,67}, whereas physostigmine increased the outflow facility. Further, intracameral injection of aceclidine, in contrast to pilocarpine, causes substantial increases in outflow facility with minimal accommodation²⁰. The aceclidine-induced facility response should be mediated by the ciliary muscle. Thus, the functions of outflow facility and accommodation can be dissociated pharmacologically or regulated by different types of receptors, as was well reviewed by Erickson-Lamy²⁰. However, as will be discussed, muscarinic receptor subtypes of the two muscle bundles seem the same in the same animal, and thus, therapeutic benefits are unlikely.

Dissociation between uveoscleral outflow and ciliary muscle relaxation

Topical PG F2 α and the isopropyl ester decrease IOP in rabbits, cats, monkeys^{24,46,50}, and humans^{3,73}, which was reviewed by Kaufman and Mittag³². The hypotensive action of PG F2 α has been attributed to an increase in uveoscleral outflow, with little effect on trabecular facility.

Morphologic study of the monkey ciliary muscle after application of PG F2 α over several days shows narrowing of the muscle bundles, with widening of the spaces and a reduction in types 1 and 3 collagen between the muscle bundles. This structural alteration may explain the observed increase in uveoscleral outflow. However, it seems unlikely that such anatomic changes could occur rapidly enough to account for the increase in uveoscleral outflow after a single topical application of PG F2 α .

Thus, a change in ciliary muscle tone^{59,72} is one possible explanation for the acute effect of PG F2 α . Results of studies on the effect of PG F2 α on ciliary muscle tone are contradictory; PG F2 α relaxed the Ach-contracted ciliary muscles of cats, rabbits, monkeys, and humans⁷². Some^{10,59,81} reported no effect of PG F2 α on ciliary muscle tone. However, PG F2 α affected the bovine ciliary muscle tone³⁷. We found no effect of PG F2 α on bovine ciliary muscle tone⁵⁹. Although the effects on ciliary muscle vary, PG F2 α increases uveoscleral outflow and antagonizes resting myopia in the monkey.

In living monkey studies, tonic accommodation averages approximately two diopters and is reversed partially by PG F2 α isopropylester (PG F2 α -IE)¹². The related ciliary muscle relaxation reduces accommodation (circular portion) and enhances uveoscleral outflow (longitudinal portion).

The ocular hypotensive effects of PGs seem to be the result of their ability to increase uveoscleral outflow. This occurs partly as a result of ciliary muscle relaxation^{5,6}, but structural changes within the muscle also contribute as stated previously³⁹. Epinephrine may partly exert its hypotensive effect via the release of PGs, because hypotension is reduced by PG synthesis inhibitors^{8,9}.

However, the effect of PGF2 α may not be caused by ciliary muscle relaxation. When the effect of the topical application of PGF2 α on refraction and accommodation was examined, topical PGF2 α reduced the IOP, but the patients' accommodation power did not change

significantly. The amplitude of accommodation was unchanged, which may suggest that IOP reduction is caused by a mechanism other than ciliary muscle relaxation⁵⁹.

It remains to be clarified whether or not endogenous PGs have a physiologic function in regulating uveoscleral outflow. We cannot deny that the PG effect may be pathophysiologic, and an increase in uveoscleral outflow may be associated with ocular inflammation, because in rabbits, the pressure reduction was inhibited by the application of indomethacin. Species differences are remarkable, because the outflow facility was increased in the bovine eye when treated with indomethacin.

Muscarinic receptor subtype and cooperative iris responses

There may be some regionalization of the accommodative and outflow functions within the ciliary muscle. Muscle fibers bundles are grouped in three major orientations, and the directional force exerted by each group differs. We should characterize such regionalization and determine whether the accommodative and outflow functions may be mediated by different subtypes of muscarinic receptor as suggested by Kaufman and Mittag³². We may some day use subtype-specific agonists to maximize outflow-mediating contraction while minimizing accommodation-mediating contraction.

The same strategy can be used to minimize pupillary constriction caused by cholinergic drugs. Induction of miosis^{19,32} is useful in the treatment of glaucoma to pull the iris root away from the trabecular meshwork. However, inducing miosis is presently disadvantageous in elderly patients with incipient cataracts, because miosis can significantly impair visual acuity.

Some muscarinic antagonists are partly selective for mydriasis as compared with cycloplegia, which suggests a difference in receptors on the two muscles. The dominant pupillary

Fig. 6. Typical responses of the three representative muscles of the bovine iris sphincter (top tracing) and the dilator (bottom tracing). The iris sphincter reacted faster than the dilator. Electrical stimulation was applied at dots: a: 0.1, 1 and 10 msec pulse widths, 3 Hz, 1.5 pulses; b: 1, 2, and 10 msec pulse widths, 5 Hz, 2.5 pulses; c: 0.5, 1, and 10 msec pulse widths, 20 Hz, 10 pulses.

constriction is via the muscarinic receptor of the iris sphincter, and the cholinergic stimulation, in turn, causes iris dilator relaxation^{60,77}. When cholinergic miosis occurs, sphincter muscle contraction and dilator relaxation work together^{31,53,55,60,63,68,77} (Fig. 6) with a different receptor subtype^{34,41}. This possibility seems beneficial for older patients who have a small pupil.

However, this is unlikely, because cholinergic contraction of the iris sphincter and cholinergic relaxation in bovine dilator muscle occur via the same subtype of the muscarinic Ach receptor⁶⁰, which was later clarified as the M3 type^{34,41,55,62,66} (Fig. 6). In an *in vitro* animal experiment, the mechanism of cholinergic contraction and relaxation of the iris is very complex, according to Takayanagi^{30,34,55,67} and Watanabe^{41,44,53}. Furthermore, it is open to question how the iris physiology is involved in regulation of IOP and outflow facility.

Muscarinic receptors around outflow channels of the iris

Furthermore, the M3 receptor predominates in isolated cat irides, radioligand studies using subtype-specific antagonists¹⁰, and Northern blot analysis using subtype-specific oligonucleotide probes²⁹. According to Zhang⁸², the human ciliary muscle expresses the mRNA of all five subtypes of muscarinic receptors. This was determined by *in situ* hybridization and/or Northern blot analysis in intact tissue samples and cultured ciliary muscle cells. Interestingly, as reviewed by Erickson-Lamy²⁰, in some individuals there may be a preponderance of m5 in the longitudinal portion of the muscle, and m3 in the circular portion of the muscle.

Muscarinic receptors are expressed in the trabecular meshwork and m3 receptors are present in cultured trabecular meshwork cells. Some expression in the ciliary muscle may be related to vascular tissue in the ciliary body, rather than to ciliary muscle cells *per se*. Studies of the monkey eye suggest that both outflow facility and accommodation responsiveness to pilocarpine are mediated by the M3 receptor^{19,21}.

The presence of muscarinic receptors, primarily of subtype M3, has been identified in

Fig. 7. Tissue culture of trabecular meshwork and smooth muscle. Left, phase contrast ; right, actin staining.

human and bovine trabecular endothelial cells⁶⁶). In addition, cells containing smooth muscle actin and myosin have been identified in the meshwork (Fig. 7). Bovine meshwork itself has contractile properties, perhaps independent of the ciliary muscle^{33,66}). Thus, it is possible to devise a therapeutically useful cholinergic that acts on the meshwork itself.

The author is studying cholinergic effects on aqueous humor outflow caused by the mechanical effects of ciliary muscle contraction⁵⁹). Muscarinic receptors are present in ciliary muscle and other anterior segment structures, but we could not find evidence for a significant increase of outflow facility *in vitro*. The physiologic roles of receptor subtypes and their distribution in the ciliary muscle and the trabecular meshwork should be clarified.

Indirectly acting cholinergic agents

Interestingly, we found that Ach (up to 10^{-5} M) alone did not contract the iris sphincter muscle⁶⁴). Ach contracts other smooth muscles from low concentrations. The degree of Ach potentiating effect by anti-cholinesterases (anti-chEs) on cardiac, skeletal, and smooth muscles varied from 1.10- to 300-fold, compared with Ach alone. The lowest threshold concentration ever reported was decreased 1,000-fold by eserine, in the case of electroplax⁵²). The present experiments showed that Ach required a 12,000 times higher concentration than carbachol (Cch) to evoke the same iris muscle contraction. Thus, the difference in potency of Ach and Cch is unexpectedly great in intraocular muscle tissue^{63,64}).

Physostigmine (eserine) and neostigmine block AchE, and prevent metabolic inactivation of Ach release from cholinergic nerve terminals. When AchE is blocked, the concentration of endogenously released Ach and its time of action is increased, as reviewed by Nardin and Zimmerman⁴³). Because the transmitter accumulates in large quantities at the neuromuscular junction, the endogenous cholinergic response is increased and prolonged⁶⁴). AntichEs have very strong cholinergic activity and a much longer duration of action in their IOP lowering ability than pilocarpine⁴²).

The use of AchE inhibitors to treat glaucoma is decreasing greatly because of their substantial adverse ocular effects⁴³). Extreme miosis occurs within half an hour of administration of eserine, and maximal IOP reduction occurs within 24 hours. Pressure reduction is extremely long-lasting, i.e., from several days to two weeks. These agents unfortunately cause iris cysts and cataract⁴³). Because AchE is present in the lens capsule, the agent increases lens hydration, resulting in lens opacity.

Anti-chE medications should be used to treat glaucoma in cases in which the patient has become intolerant of pilocarpine. The chE inhibitor is very effective in glaucoma of aphakia or pseudophakia⁵¹). In uncontrolled glaucoma treated with pilocarpine or carbachol, changing the miotic therapy to echthiophate iodide improved pressure in 60% of the eyes⁴³). Unfortunately, the drug cannot be used in younger patients, because of the degree of miosis and accommodative spasm.

Beta agonists and ciliary muscle tone

β -adrenergic agonists are considered to increase the outflow facility via activation of the β -receptors. To our best knowledge, only one study that used organ culture eyes²²⁾, showed increased outflow facility during the perfusion of epinephrine. However, there is no report in the literature that β -agonists increase the outflow facility in enucleated animal eyes⁶⁶⁾. More interestingly, the outflow facility rather decreased by increasing the concentration up to 10^{-3} M. No report has shown that the trabecular meshwork is innervated, although slight staining of peptides and purines^{56,71)} is evident.

Adrenergic nerve stimulation decreases the outflow facility in monkeys but has no effect on uveoscleral outflow. Interestingly, adrenergic agonists do not mimic the effects of adrenergic nerve stimulation. Adrenergic drugs^{42,49)} may directly affect the trabecular meshwork and the uveoscleral outflow, perhaps via release of prostaglandins^{9,61)}.

Phosphodiesterase (PDE) inhibitors, ciliary muscle, and outflow channels

Regarding adrenergic effects, many other inconsistencies exist in the field of IOP regulation and related intraocular muscle study. How do adrenergic agonists and antagonists lower IOP? To our knowledge, few experiments using enucleated animal eyes have been carried out and the mechanism of IOP reduction is open to question despite many experiments using living animals. Why is the adrenergic nerve supply scarce in the human trabecular meshwork, although topical epinephrine has long been used clinically? Does this mean only that the trabecular meshwork has β receptors that are not related to adrenergic innervation? If adrenergic effects occur via release of PGs, thereby decreasing IOP, then does adrenergic influence on IOP reduction occur via uveoscleral route?

Furthermore, why is adrenergic relaxation of the ciliary muscle small or even nil⁵⁹⁾ (Fig. 8),

Fig. 8. Effects of epinephrine and isoproterenol on nerve-mediated contractions (a, b) and carbachol-induced precontraction (c) of the bovine ciliary muscle.

Table 3. Effects of β agonists and PDE inhibitors on Ciliary Muscle, Outflow Facility and Cell Shape Change of Trabecular Meshwork

	β_1 agonists		β_2 agonists	β agonists		PDE inhibitors
	dobutamine		salbutamol	epinephrine	isoproterenol	theophylline IBMX, caffeine zaprinst
ciliary muscle: tone	→	↘	→	→	↘	↓
ciliary muscle: nerve-mediated contraction	→	↘	→	→	↘	↓
cell shape change	++		—	+		+++
outflow facility	→	↘*	→	→	↘*	↑↑

PDE: phosphodiesterase

although most smooth muscles are reciprocally innervated and the ciliary muscle greatly relaxes with PDE inhibitors. Theophylline inhibited the nerve-mediated contraction of the ciliary muscle, and also reduced the tension of the pre-contracted bovine ciliary muscle by carbachol³⁵. In contrast, adrenergic drugs (epinephrine, isoproterenol, dobutamine, and salbutamol) had only a small effect on the ciliary muscle and the outflow facility of the eye⁶⁶.

It is beyond the scope of this brief review to describe the effects of drugs on the intraocular smooth muscle tone, nerve-mediated contractions, and cell shape changes of the smooth muscle, or the trabecular meshwork and to search for the physiologic and clinical meanings. Only an example is provided in Table 3. Why do PDE inhibitors play a much greater role than adrenergic agonists^{35,66,74,75}? Does this mean that an intracellular increase of cGMP occurs as a result of the application of PDE inhibitors, including zaprinast, in addition to the increased cAMP concentration? Species differences make these problems extremely difficult to solve, but they should be addressed in the future.

In considering the effects on IOP, each of these transmitters or modulators may have ocular sites of action and may induce significant net changes in IOP that do not necessarily reflect their potential effects on individual intraocular muscles.

Multiple factors should influence aqueous humor outflow, IOP, and intraocular muscle function. The narrow range and stability of normal IOP suggest that this system somehow is regulated⁵⁸, although few data are available on the metabolic control of outflow facility⁶⁵. A possible regulatory system probably incorporates a feedback loop consisting of a sensory or afferent pathway and an efferent pathway through the autonomic nervous system. It should be clarified how the IOP, aqueous outflow rate, and intraocular muscles are moderated. The discoveries of neuropeptides and purines, the effects on outflow facility and intraocular muscles, and the association of cell shape change, intraocular muscle contraction with outflow facility, and especially animal species differences, all indicate a previously unappreciated heterogeneity and complexity of the ocular innervation and the mechanisms governing IOP and intraocular muscle physiology^{53,59,68}.

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