Attempts at reverse perfusion of the trabecular meshwork in different monkey species
(Cercopithecus ethiops, Macaca mulatta, and Macaca speciosa)

Reinhard Dannheim and Ernst H. Bárány

A small plastic bell connected to a reservoir suspended on a strain gauge and to a pressure recorder was fixed over a fistula in the scleral wall of Schlemm’s canal. The anterior chamber was similarly connected to a reservoir and a pressure transducer. In a majority of vervet eyes an increase of the pressure in the bell over the intraocular pressure caused a bulk inflow into the canal through the bell and a reflux from the anterior chamber to its reservoir. Thus reverse perfusion of the trabecular meshwork from the inflated canal took place. Conventional facilities seen from the anterior chamber increased with increasing pressure in the bell. Analysis of the data indicate that the increase in facility is due to several factors. In the inflated region scleral resistance is short-circuited. Probably inflation of the canal also increases facility by reducing the area of normally collapsed regions of the canal where little filtration can take place. Moreover, a gradual change in size of the inflated region depending on the pressure relations between anterior chamber and canal appears as a facility during reverse perfusion. In the rhesus monkey the inflow from the bell became fast at high pressure levels, but no detectable reverse perfusion took place and the facility was not changed. The reason for this behavior is not quite clear. In the one stump-tail eye tested, no inflow from the bell was achieved.

In order to measure the real resistance of the trabecular meshwork in monkey eyes we had hoped to be able to perfuse it in the reverse direction. A fistula in the scleral wall of Schlemm’s canal would be used as a means of introducing fluid in measured amounts and under known pressure. However, we ran into unexpected difficulties. In a preceding paper observations in eyes with a fistula joining Schlemm’s canal to the ambient air are described. The present paper deals with experiments with controlled pressure in the fistula. Both series of experiments indicate that under certain conditions the canal is partly collapsed.

Methods

A total of 40 vervets (Cercopithecus ethiops), 3 rhesus monkeys (Macaca mulatta), and 1 Thailand stump-tail (Macaca speciosa) were used once the technique of fixing the bell to the sclera was mastered. The monkeys were of either sex, the vervets varying between 1.8 and 6.3 kilograms and the rhesus monkeys between 1.8 and 2.5 kilograms, while the single Thailand stump-tail was about 3 kilograms. Anesthesia was induced by 10 mg. of methohexital sodium (Brevital, Eli Lilly

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and Company) per kilogram intramuscularly and then pentobarbital sodium was given intravenously up to 25 to 30 mg. per kilogram.

**Operative procedure.** The lids were held open with sutures and a canthotomy was made. The upper aspect of the conjunctiva was opened about 5 mm. from the limbus over half the circumference and bluntly dissected from the sclera. Holding sutures were placed in the insertions of the three upper recti. A lamellar dissection of the cornea extending about ½ corneal diameter beyond the limbus was then performed (Fig. 1, a). Bleeding limbal vessels extending for 5 or 6 mm. in either direction from the planned fistula close to 12 o'clock were cauterized with a hot needle. A piece of smooth polyvinylchloride (PVC) film, 0.2 mm. thick and about 7 mm. in diameter (Fig. 1, b), was then glued to the eye. Some Eastman 910 Adhesive (we had only the industrial version) was smeared over the piece of film, which was then pressed against the slightly moistened limbus region. The adhesive polymerizes in a few minutes but we waited 10 minutes before proceeding. With pieces of a razor blade, a window approximately 1.0 by 1.5 mm., with its long axis in the anteroposterior direction, was then cut in the film and the film removed over this area. While the scleral tissue was gripped within the window area, a cut was then made along one long side of the window, at the same time as the scleral tissue was pulled outward and away from the incision. The operation was performed under a binocular operation microscope with an extra microscope for the assistant, under x16 to x40 magnification. One can clearly see when the incision comes close to the canal. In the vervet the region of the canal appears as a gray band between the clear cornea and the white sclera. When it had been identified, the canal itself was opened while one pulled at the sclera. The canal could then be seen as a dark cleft which widened as one pulled. Aqueous humor filled the gap. Only then were the remaining incisions made into the sclera and the scleral substance removed during continuous traction to keep the canal open. All tissue covering the canal must be carefully removed so that the trabecular meshwork (Fig. 1, d) is lying directly at the floor of the pit in the sclera.

In the rhesus monkey the procedure started in the same manner but there the region of the canal is not gray but violet, because of a collection of vessels overlaying the canal. Often this band of vessels was visible already before the dissection had proceeded very far. These vessels are unavoidably cut when the canal is opened and give rise to considerable hemorrhage. Attempts to prevent or stop the bleeding by the use of Octapressin (Sandoz Pharmaceuticals, Hanover, N. J.) were unsuccessful.

Sometimes there was also some hemorrhage in the vervet but much less and much less disturbing for the conduct of the experiment. Nor was bleeding a problem in the one stump-tailed operated on. Besides a hemorrhage, the presence of unexpanded adherent can cause complications. As Eastman 910 spreads easily over moist surfaces before it polymerizes it is necessary to wait 10...
minutes after the PVC film has been put in place before the region of the canal is approached. Otherwise adhesive might spread over the window in the canal and obliterate the fistula. After the canal had been opened a ring of hard PVC (Fig. 1, e) surrounding the window was glued to the film with the aid of vinyl cement (Testors vinyl cement). The inner diameter of the ring was 1.5 mm, the outer 4.0 mm. In order for the ring to be attached, the surface of the film has to be kept dry with the aid of fine wicks which collect the aqueous as it emerges from the fistula. After a last check that the opening to the canal was not blocked by glue or coagulated blood, the bell (Fig. 1, f) machined out of hard PVC was glued to the ring, again with vinyl cement. The bell had two holes in the top through which two narrow and soft polyvinyl tubes entered its lumen (Fig. 1, g). As soon as the bell was in place it was filled with heparin solution in saline (5,000 units per milliliter, only in vervets) in order to prevent coagulation. The perfusion fluid in the reservoirs also contained heparin, 500 units per milliliter. (In some experiments we have given systemic heparin intravenously, 1,000 units per kilogram as a priming dose and then an infusion of 1,000 units per kilogram per hour. This, however, sometimes led to considerable bleeding into the bell.) After the bell was in place and had been tested for absence of leaks, two needles were fired into the anterior chamber and connected with a perfusion and recording system as described. Fig. 3 shows the various connections when the preparation was complete.

Results and discussion

I. Pressure measurements. Fig. 2 shows the pressures in the bell and in the anterior chamber in 15 vervet eyes in which no considerable bleeding was visible. In none of them was there any leak. Pressures were recorded before the reservoirs had been connected to the bell and the anterior chamber. All holding sutures were completely relaxed. The values refer to the first two or three minutes of recording only and possibly are not real steady-state pressures, although the records were reasonably stable. Evidently there is a considerable variation from animal to animal in both pressures. The average (open circle in Fig. 2) in the anterior chamber is 16.6 mm. Hg and in the bell is 13.2. The average of the individual differences between anterior chamber and bell is 3.44 ± 1.56 mm. Hg (S.E.M.), P < 0.05.

The pressure in the bell is a resultant of the pressure in the canal and in the open vessels of the sclera. It is hard to see how it could be an underestimate. Therefore our results are in partial disagreement with those of Perkins and Sears. They found a very small pressure difference between the canal itself and the anterior chamber. Both these authors pushed a tube into the canal through a slit in the sclera. It seems possible that some damage could have been done to the endothelium of the inner wall of the canal, considered to be the site of the outflow resistance. On the other hand, low pressure differences were sometimes seen in our experiments too.

In the rhesus monkeys, bleeding was so disturbing that no reliable results were obtained. Pressure in the bell often reached 30 to 40 mm. Hg. Evidently there are arterioles in the vascular bundle which has to be incised to open the canal.

Pressure in the single stumptail eye was 20.8 in the anterior chamber and 13.4 mm. Hg in the bell.

II. Flow measurements and facility at different pressures. If the reservoirs are

Fig. 2. Intraocular pressure, $p_a$, and pressure in the bell, $p_b$, both in millimeters of Hg. Each dot ($\bullet$) represents one vervet eye. The circle (O) denotes the average. The diagonal line connects equal values of $p_a$ and $p_b$. 

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connected to the eye the arrangement is that of Fig. 3. The plastic bell is connected to reservoir B and to a pressure transducer recording $p_B$. Flow from the bell to its reservoir is called $B$ and its sign is counted positive when the flow goes into the reservoir (to make the symbol consistent with that in reference 1). The pressure in the anterior chamber is $p_i$ and the rate of flow from its reservoir is $Q$. Thus it is counted positive when fluid runs into the anterior chamber. By adjustment of the positions of the reservoirs B and Q the two pressures $p_B$ and $p_i$ can be set at will. One sequence of pressures $p_B$ and $p_i$ used is shown at the top if Fig. 4, others in Fig. 10. If one leaves $p_B$ unchanged and lets $p_i$ move between two levels, one obtains a conventional facility from the change in pressure ($\Delta p_i$) and the resultant change ($\Delta Q$) in inflow rate ($Q$) into the anterior chamber. This facility obtained by the change in intraocular pressure ($p_i$) will for the present purposes be called $C_i$.

$$C_i = \frac{\Delta Q}{\Delta p_i}.$$  

For reasons explained elsewhere$^6$ one should average $C_i$ obtained from the rise in pressure with that obtained from the decrease in pressure; this has been done throughout.

**Description and analysis of 2 vervet experiments.** The effect on $C_i$ of a stepwise increase and then decrease in pressure $p_B$...

![Diagram of experimental arrangement](image_url)

**Fig. 3. Diagram of the experimental arrangement.** Note that flow is counted positive when it goes to reservoir $B$ and when it leaves reservoir $Q$.

![Graph of experiment](image_url)

**Fig. 4. Time course of an experiment.** Top, broken lines = intraocular pressure, $p_i$, in millimeters of Hg; continuous lines = pressure in the bell, $p_B$, in millimeters of Hg. Numbers represent periods of $p_B$ and correspond to points in Fig. 5. Bottom, conventional facility, $C_i$, in microliters per minute per millimeters of Hg.
is illustrated in Fig. 4. C, which starts with the value 1.6 μL per minute per millimeter of Hg, at first changes but little as \( p_B \) is increased. But when \( p_B \) exceeds \( p_i \) a considerable increase in facility is seen, with a maximum reading of 3.3 units. When \( p_B \) is lowered again, \( C_i \) decreases to nearly starting levels. Fig. 5 illustrates the flow from the reservoir B and from the reservoir Q as a function of the pressure in the bell, \( p_B \). The left half refers to the situation with the Q reservoir at its lower level and the right half to that with the Q reservoir at its upper level. Since there are two periods of low pressure to every high pressure period, the values obtained with the two low pressure ones have been averaged. The common abscissa is pressure in the bell, \( p_B \).

At the top the differential between intraocular pressure, \( p_i \), and \( p_B \) is plotted. It is not a straight line. Because of needle resistance \( p_i \) is not completely independent of the amount of fluid flowing in and out of the anterior chamber and therefore it rises somewhat when \( p_B \) is increased.

The figure shows that \( Q_i \), the rate of flow from the reservoir into the anterior chamber, undergoes but little changes while \( p_i-p_B \) is positive and also when it is moderately negative, but when \( p_B \) becomes considerably higher than \( p_i \) a backflow

![Figure 5](http://iovs.arvojournals.org/pdfaccess.ashx?url=/data/journals/iovs/932999/)

Fig. 5. Effects of varying the pressure in the bell, \( p_B \), in the vervet monkey (same experiment as Fig. 4). The left half shows values obtained with the intraocular pressure, \( p_i \), at its lower level, the right half refers to the higher level of \( p_i \). The difference between these levels was 4 to 5 millimeters of Hg. The numbers of certain points in the \( Q \) graph correspond to the same periods in Fig. 4.
from the anterior chamber into the reservoir Q occurs. (This backflow is much larger in the left than in the right graph.) When pressure \( p_B \) is again stepwise reduced, the backflow disappears. The maximum value of the backflow is more than 15 \( \mu L \) per minute (in the left graph) and thus greatly exceeds any possible rate of secretion. Evidently fluid has been forced from the bell into the anterior chamber. Turning now to the graph of B, it was mentioned that the rate of flow is counted positive when fluid leaves the eye and goes into the bell. At the very start of the experiment, with \( p_B \) low, this is seen. But even at a \( p_B \) of 17 to 18 mm. Hg, which is about 4 to 5 mm. above intraocular pressure, in the lefthand graph, B is still positive and there is no inflow from the bell into the eye. With a further increase in pressure, however, the flow from the bell into the eye becomes quite rapid and B becomes negative. At 26 mm. Hg some 80 to 90 \( \mu L \) per minute go into the eye. When the pressure is lowered again the flow through the bell decreases and if the curve is extrapolated linearly, as shown by the broken line in the left graph, it seems to reach zero at approximately 13 mm. Hg, where \( p_l = p_o \). Further down, \( p_B \) again is too low to force fluid into the eye; on the contrary, some flow into the bell occurs from the eye and B is positive.

In the right-hand graph it is seen that the shift from negative to positive B occurs quite close to \( p_B = p_l \). The course of facility \( C_l \) is shown at the bottom of the figure. The same values of \( C_l \) appear on both sides but at slightly different \( p_B \), because \( p_B \) is influenced a little by \( p_l \).

It is seen that changes in \( p_B \) at the lower end have no influence but that elevation of \( p_B \) above \( p_l \) causes a rise in \( C_l \). When \( p_B \) is lowered again, \( C_l \) returns but by another route, indicating the presence of a hysteresis element.

We will now attempt an interpretation of the results shown in Figs. 4 and 5. The first observation to be considered is the fact that inflow from the reservoir Q is hardly affected when \( p_B \) is increased from 5 to 10 mm. at an intraocular pressure (\( p_i \)) of 13 mm. Hg. This was so during the rising as well as during the falling phase. In the preceding paper it was argued that when pressure in the fistula is low the trabecular meshwork will be pressed against the scleral wall of the canal and free communication between the fistula and the remaining part of the canal prevented. Undoubtedly a similar mechanism was present at the low values of \( p_B \). Even increasing \( p_B \) to 18.5 mm., well above intraocular pressure, had little or no effect on Q and B. If a collapse of the meshwork was present it seems that the inner wall of the canal stuck to the outer wall despite the 5 mm. excess pressure acting from the fistula. Further increase of \( p_B \) to about 22 mm. suddenly opened the canal to the fluid from the bell and a rapid flow into the canal from the bell took place. Part of this flow passed through the trabecular meshwork in a reverse direction, causing the considerable negative value of \(-10\) in Q, but the main part must have passed out through scleral channels since B was in fact 54 \( \mu L \) per minute (point 4, left graph, Fig. 5). Further increase in \( p_B \) caused more fluid to go backward through the trabecular meshwork but even larger quantities to go through scleral channels. On the downward leg of the experiment the course of flow B differs from the rising leg (left-hand graph). The significance of this observation is doubtful. The slope was rather constant between \( p_B = 26 \) and \( p_B = 17.5 \) mm. and when extrapolated it strikes the abscissa at a pressure very close to the intraocular one where the connection between the fistula and the canal would be expected to fail again (see dashed line, Fig. 5). We have no point in this region at low \( p_l \) but it seems probable that the B curve would have had a course not very far from the broken line if points had been available. The course of the B curve at high \( p_l \) (right-hand graph) confirms this.

The course of Q at high \( p_l \) differs
Fig. 6. Time course of an experiment. (For explanation, see Fig. 4.)

Fig. 7. Effect of varying $p_s$ in the vervet monkey (same experiment as Fig. 6). (For explanation, see Fig. 5.)
markedly from that at low $p_1$. It is seen that even with $p_B = 27$, exceeding $p_1$ by 8 mm., the reverse flow into the $Q$ reservoir was only 2.5 $\mu$L per minute. The corresponding value with $p_B = 26$ and $(p_1 - p_B)$ lower by 3.5 mm. Hg, left-hand graph, is 15 $\mu$L per minute. This finds its expression in the large facility $C_1$ at the highest $p_B$; $C_1$ is of course calculated exactly from differences in $Q$ between a higher and a lower $p_1$. Facility $C_1$ is plotted at the bottom of the graphs. It is evident that as the connection between the fistula and the canal opens, higher values of facility occur. A hysteresis loop would be expected if stickiness plays a part but it should run in the opposite direction. The reason for the present loop is not clear.

Figs. 6 and 7 depict another vervet experiment with the same protocol. The principal features are quite the same as in Figs. 4 and 5 but the values of $Q$ span over a wider range. $B$ starts to become negative close to $p_B = p_1$. At a low $p_B$, $Q$ is strongly positive, and at higher $p_B$ much more negative than in the first experiment (Figs. 4 and 5). The shape of the $C_1$ curve is similar but reaches higher values. The hysteresis loop again is in the wrong direction to be explained by stickiness.

**Reasons for the dependence of $C_1$ on pressure in the fistula.** There are several possible mechanisms which can come into play simultaneously:

1. Increase in size of the relevant pores in the trabecular meshwork when the canal is inflated.

2. A "Sears mechanism." Sears has pointed out that the canal could in fact be a cleft with only a virtual space. If this were so, one would expect the collapsed parts to contribute little to filtration. Opening up the canal by means of an increasing $p_B$ might increase filtration area. This could be an important effect especially in the vervet, where the scleral spur is almost absent and the canal very shallow.

3. The "Weekers mechanism." Weekers and colleagues suggested that compression of the trabecular meshwork by the intraocular pressure could decrease facility. This effect would counteract the increase in flow with intraocular pressure when flow over the meshwork is in the normal, forward direction, but it would augment the decrease in flow caused by a rise in intraocular pressure when flow is in the reverse direction. Thus, in reverse filtration, it would increase $C_1$.

4. Short-circuiting of scleral resistance by the bell. If part of the canal is in open communication with the bell and the bell in open communication with its reservoir by way of a low resistance connection, then the bell and its connection short-circuit the scleral resistance. Increase in aqueous flow from the anterior chamber through the meshwork into this part of the canal causes but little increase in pressure due to the short circuit and thus the apparent resistance of the short-circuited sector as seen from the anterior chamber is lower than under normal conditions. Conversely, if $p_1$ is lowered and influx from the canal increased, the pressure in the canal decreases less because of the short circuit, the change in influx becomes larger, and facility appears augmented.

5. Reversed flow through the meshwork might be easier than forward flow. This is only a remote possibility and we do not believe in it.

6. Pressure-dependence of the fraction of trabecular meshwork engaged in reverse filtration. This, we think, is an important factor. Fig. 8 has been constructed to explain it. In Fig. 8, $I$ the situation with $p_1 > p_B$ is illustrated. As mentioned before, there will be a collapse of the meshwork close to the fistula. All movement of fluid across the meshwork is in the forward direction, from the anterior chamber, and total outflow is large (broad arrow $F$, Fig. 8).

In Fig. 8, $II$ the $B$ reservoir is a little higher than the $Q$ reservoir. Some reverse filtration across the meshwork takes place near the bell, but over most of the circumference flow is in the forward direction. $Q$ therefore still is strongly positive, total out-
Fig. 8. Flow situation at different combinations of pressure in the fistula and the anterior chamber. Reservoir \( Q \) determines \( p_i \), reservoir \( B \), \( p_B \). Arrow \( F \) indicates conventional outflow of aqueous. \( N \) indicates the position of the neutral zone.

Flow is somewhat reduced but \( F \) is still considerable. This situation is in fact observed in points 3 and 7 of Fig. 5 right, point 8 of Fig. 7 left, and especially points 4, 7, and 8 of Fig. 7 right.

In Fig. 8, III the pressure in the bell has been increased further. Reverse filtration now takes place over an extended area and forward flow (\( F \)) is correspondingly reduced. The net effect now is a negative \( Q \), as observed at the higher values of \( p_B \) in Figs. 5 and 7.

Why does reverse filtration not occur over the whole circumference at once when \( p_B \) exceeds \( p_i \) (as indeed we had expected it to do at the start of our experiments)? The reason must be that a pressure drop occurs along the canal. This drop is caused by the longitudinal resistance of the canal in conjunction with the facilities toward both sides, into the anterior chamber, and out through the collector channels. Therefore, with moderate excess of \( p_B \) over \( p_i \), a neutral zone \( N \), where no filtration across
the meshwork takes place, is reached as one moves away from the fistula. Closer to the fistula the canal is inflated, filtration is reversed; beyond the neutral zone filtration is in the normal direction and the canal is more or less a cleft with reduced filtration area. When intraocular pressure is changed, the neutral zone shifts its position. With \( p_i \) high, \( N \) is closer to the bell (Fig. 8, III), with \( p_i \) low it is further away (Fig. 8, IV). This movement of the neutral zone appears as an added facility seen from the anterior chamber. Facility is pressure-dependence of filtration. With this mechanism filtration changes not only in proportion to pressure but more, since pressure alters the conditions for filtration. In the present system conditions for reverse filtration improve for two reasons when \( p_i \) decreases: the increased pressure head, \( p_n-p_i \), and the increased surface over which the canal is fully opened; and filtration is correspondingly enhanced by the mechanisms enumerated in the earlier paragraphs.

Static facilities. The mechanism described implies a nonlinear relation between \( Q \) and \( p_n-p_i \). In Figs. 5 and 7, \( Q \) is plotted against \( p_n \) but since \( p_i \) differed only little from one end of the \( p_n \) scale to the other, the curves can as well be read as showing the dependence of \( Q \) on \( p_i-p_n \). The nonlinearity is evident.

The fact that the nonlinearity is present makes the use of the conventional differential facility \( C_i = \frac{\Delta Q}{\Delta p_i} \) questionable. If the functional relation between \( Q \) and \( p_i \) is curved, such a \( C_i \) does not represent the slope correctly, \(^9 \text{,}^{10} \) But more importantly, such a \( C_i \) is a poor measure of the state of the meshwork.

One could try to use a static facility by dividing \( Q \) with the appropriate driving pressure. But what is the appropriate driving pressure? We only know \( p_n \) but we have no certainty that it prevails all around the canal; on the contrary, it is probable that pressure drops off as one moves away from the bell. By using \( p_n-p_i \) as the pressure head one therefore overestimates the pressure and underestimates the facility. A further unknown is the rate of secretion of aqueous which in itself causes a negative contribution to \( Q \). With high \( p_n \) and large negative \( Q \) the rate of secretion (somewhere around 1 to 3 \( \mu \)L per minute) has little importance. But when the absolute \( Q \) value is low it really matters. This greatly hampers the use of static facilities. Nonetheless we have calculated them for those points in Figs. 5 and 7 where negative \( Q \) is at least 6, where undoubted reverse filtration occurred. For comparison we have calculated these static facilities for the two alternatives of no secretion and 3 \( \mu \)L per minute secretion. The data are presented in Table I. Columns 3 and 4 show that points 4 and 5 of Fig. 5 left have similar static facilities, around 1 unit, while static facilities in points 4, 5, and 6 of Fig. 7, left, are around 2.5 to 3. The fact that similar static facilities have been found at several levels of \( p_n \) in vervet 3,655 means that no change in the conditions for filtration did occur over the pressure range in question. This could be due to either complete opening up of the canal already at the lower \( p_n \) or stickiness between the walls. Total opening up of the canal would be expected to yield static facilities at least around 3 as observed in vervet 3,654 (columns 3 and 4). Therefore it seems likely that the low and constant static facility in Case 3635 was due to sticking between the two walls of the canal.

Since intraocular pressure tends to decrease the open part of the canal one would expect static facility to decrease with \( p_i \). This is seen in columns 7 and 8 with regard to vervet 3,654.

The table also shows some data for the only point in 5 rhesus eyes where \( Q \) was negative enough by our criterion and a static facility could be measured. Surprisingly, static facility was higher at the higher intraocular pressure. As discussed in the next paragraph the rhesus eye behaved very differently from the eye of the vervet.

Experiments in the rhesus monkey. We have tried to perform the same experiment
Table I. Static facility $C = Q/(p_i - p_B)$ in 3 experiments

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Point</th>
<th>1.</th>
<th>2.</th>
<th>3.</th>
<th>4.</th>
<th>5.</th>
<th>6.</th>
<th>7.</th>
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<td>1.06</td>
<td></td>
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<tr>
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<td>(Fig. 7)</td>
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<td>-10.8</td>
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<td>1.61</td>
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</table>

Subscript L (columns 1 to 4) refers to the lower $p_i$ and subscript H (columns 5 to 8) to the higher $p_i$. The additional subscript S means that a secretion rate of 3 microliters per minute has been assumed. $Q$ is in microliters per minute, $p_i$ and $p_B$ in millimeters of Hg, $C$ in microliters per minute per millimeter of Hg.

in 6 eyes of rhesus monkeys. In one the meshwork was damaged but in 5 the experiment was technically successful except for the bleeding mentioned under Methods. The results were all very similar and differed markedly from those in the vervet. Fig. 9 shows one of these experiments. The increase in $-B$, flow from the bell, in these monkeys appeared at a higher $p_B$ than in the vervet, at values where $p_B$ exceeded $p_i$ by 5 to 10 mm. Hg. This was so on the ascending as well as on the descending leg and the $p_B$ at which the increase in $-B$ occurred was the same at high or at low $p_i$. All this is in contrast to the vervet where flow from the bell started as soon as $p_B$ exceeds $p_i$ by a small amount (with the exception of the one point in Fig. 5 left, ascending leg). It seems that something other than the collapse of the meshwork around the fistula hampered inflow from the bell in the rhesus experiments. There was at most a few microliters per minute of reverse filtration (under the assumption of no secretion) and this filtration changed hardly or not at all with $p_B$ despite the
fact that \( p_B \) was up to 8 to 10 mm Hg in excess of \( p_i \). This was true in all 5 eyes. Facility \( C_i \) was low and failed to show the marked increase at high \( p_B \) seen in the vervet.

All these observations taken together indicate that in this species we were unable to control the pressure in the canal by means of the bell and to perfuse any appreciable fraction of the meshwork in the reverse direction. It seems that the flow from the bell, which appeared at a high \( p_B \) and independently of \( p_i \), was not into the canal but into the vessels of the sclera. We do not know why the rhesus eye differed so markedly from the vervet eye. Possibly the heavy bleeding blocked the canal in spite of systemic and topical heparin. It could also be that the Eastman 910 Adhesive in this species penetrated the sclera to a larger extent than in the vervet and cut off the canal. Finally it is just barely possible that the rhesus canal is in fact segmented into short portions and does not allow longitudinal flow over considerable fractions of its total length. We are not aware of any anatomical data on this point.

**Experiments with different protocols.**

Beside the experiments illustrated, a large number of others were made with several different protocols which are shown schematically in Fig. 10. Our intention was to compare \( C \) values derived from changes in \( p_i \) with values derived from changes in \( p_B \). This turned out not to be possible because \( p_B \) did not penetrate far enough into the canal. However, information can be derived also from some of these experiments, which are summarized in Table II. The table shows the changes in \( B \), in \( Q \), and in \( C_i \) (where present) caused by the change in \( p_B \). The periods utilized in the calculations for the table are marked by dots (for \( B \) and \( Q \)) and by arrows (for \( C_i \)) in Fig. 10. Periods with identical pressure relations were averaged.

It is seen that in 9 of 15 vervet eyes there was a change in \( Q \) in the same direction as in \( B \) when \( p_B \) was varied. In 6 unresponsive vervets (marked with U in the Comments column) there was virtually no change in \( Q \) and only very small changes in \( B \). \( C_i \) increased in those where \( Q \) changed, but hardly or not at all in the others. This is in agreement with the two experiments described extensively above and heading the table. In rhesus monkeys, very little change in \( Q \) or \( C_i \) occurred despite large changes in \( B \). Little clear-cut increase in \( C_i \) could be discerned. In the stumptail very little change in \( B \) and none in \( Q \) or \( C_i \) was seen.

Pilocarpine systemically after pretreatment with a small dose of atropine was tried in 3 vervet experiments. The other eyes of the same animals were first run without drugs. Two of them were unresponsive without drugs and one stayed unresponsive after drugs. Comparison with the untreated eye shows that in all cases the pilocarpine effect on starting facility was weak, and in the one that stayed unresponsive it was absent or maybe even negative. Thus the effect of pilocarpine is questionable and more experiments are needed.
Table II. Effects of changes in $p_B$ on $B$, $Q$, and $C_i$ in different species

<table>
<thead>
<tr>
<th>Experiment No.</th>
<th>Type</th>
<th>$p_B$ Change</th>
<th>$C_i$ Change</th>
<th>Comments</th>
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<td>$B$</td>
<td>$Q$</td>
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<td>From</td>
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$B$ and $Q$ are in microliters per minute, $p_B$ in millimeters of Hg, $C_i$ in microliters per minute per millimeter of Hg.

The Type column refers to Fig. 10. A U. in the Comments column indicates unresponsive vervet eyes.

In experiments 3684*, 3686*, and 3687* the animals were given 0.05 mg. per kilogram of atropine sulfate intravenously 1 hour before the experiment and 2 mg. per kilogram pilocarpine hydrochloride intramuscularly 15 minutes before the perfusion.

The results in the rhesus, the stumptail, and in the unresponsive vervets indicate that in these cases we were unable to reach into the canal with the pressure, $p_B$, employed. Possible reasons for this failure have been discussed previously. It can be mentioned that in one vervet experiment, where two fistulas were made, 180 degrees apart, and two bells successfully placed in the same eye, we were unable to affect $B$ in the one bell by increasing $p_B$ in the other. Thus, at least in this case, there was no free communication around the canal with the pressure range used. We have not raised the pressure above 35 mm. Hg.

**Comments**

The findings in the present paper have a bearing on the mode of action of pilocarpine on outflow resistance in the vervet monkey. As pointed out, it is hard to see how ciliary muscle contraction could affect pore size as such. It was therefore proposed that contraction changed the effective area of filtration by separating trabecular lamellae and allowing the aqueous access to a larger part of the inner wall of the canal. If the canal is in fact partly collapsed under normal pressure conditions, as our experiments seem to indicate, one would expect lessened filtration through those parts of the inner wall which touch the outer wall. The large improvement in outflow facility caused by ciliary muscle contraction in this species conceivably could then be due to a separation of the two walls of the canal in the previously collapsed regions. This would be a way of...
modulating effective filtering area. Our results do not allow any decision as to whether the marked difference in pilocarpine effect on outflow resistance between different monkey species is due to the anatomical differences determining the degree of collapse of the canal.

We are very grateful to Mr. Folke Högberg for the precision machining of the rings and bells and to Mrs. Malin Svensson and Mrs. Ingalill Wersäll for their customary expert assistance with the experiments.

REFERENCES