Original Articles

The filtration coefficient of the intraocular vasculature as measured by low-pressure perfusion in a primate eye

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Anterior chamber perfusion of a series of rhesus monkey eyes was carried out at intraocular pressure levels above and below episcleral venous pressure. For intraocular pressures above venous pressure the slope of the flow-pressure curve was a measure of total facility of the eye. The average value for total facility was found to be 0.66 ± 0.11 μL min.$^{-1}$ mm. Hg^{-1} which is in good agreement with previously published data for the rhesus monkeys similarly anesthetized. The slope of the flow-pressure curve at intraocular pressures below venous pressure was a measure of the filtration coefficient of the intraocular vascular tree. The filtration coefficient determined by this technique was found to be 0.30 ± 0.06 μL min.$^{-1}$ mm. Hg^{-1}. These data are in good agreement with the value for the filtration coefficient determined previously using osmotic transient experiments in the same species.

Key words: blood-aqueous barrier, facility of outflow, filtration coefficient, rhesus monkey eye, anterior chamber, intraocular pressure, episcleral venous pressure.

The filtration coefficient of the blood-tissue barrier of the eye is a measure of the volume flux which can occur between the intravascular and the extravascular compartment across the endothelial wall of the intraocular vascular tree. This passive flux across the endothelial cell occurs as a result of osmotic and hydrostatic gradients and contributes to some extent to the formation and drainage of extravascular fluids within the eye.[1]

The filtration coefficient of the blood-tissue barrier in a primate has been estimated by measuring the flux occurring across the vascular wall as a result of transient changes in osmotic pressure of the blood.[2] The purpose of this experiment was to calculate the filtration coefficient of the whole eye by measuring the flux occurring across the vascular wall as a result of transient changes in hydrostatic pressure.

Methods and materials

Preparation of animals. Nine normal rhesus monkeys, averaging 3.4 kilograms were studied. In one animal, only one eye was used so that

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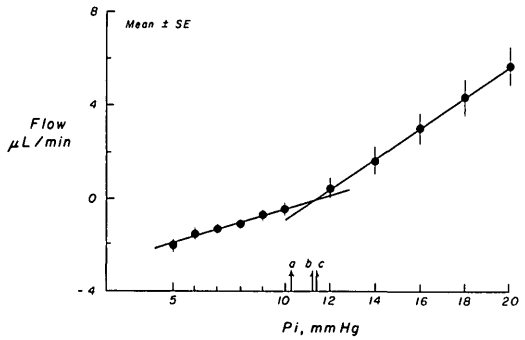

Fig. 1. Flow into eye from reservoir for different reservoir heights (millimeters of Hg) (means of 17 eyes). A = angular venous pressure, mean of 7 animals. B = apparent discontinuity in flow diagram below which trabecular outflow assumed to be 0. C = calculated undisturbed intraocular pressure under anesthesia, all animals.

a total of 17 eyes were studied. Each animal was tranquilized with intramuscular phencyclidine 2.2 mg. per kilogram and anesthetized with intraperitoneal pentobarbital 15 mg. per kilogram. The animals were placed in the supine position and allowed to breathe room air through an endotracheal tube. Each anterior chamber was cannulated with a No. 25 disposable needle, and the eye perfused with TC199 (Tissue Culture Medium 199, Grand Island Biological Company, Chagrin Falls, Ohio) as described previously.[3] Evaporation from the cornea was prevented by covering the surface of the eye with a small piece of Saran Wrap.

Experimental procedure. A constant pressure perfusion reservoir was set at a level of 10 mm. Hg for each eye and allowed to remain there until the flow into the reservoir or out of the reservoir was constant. In one eye the intraocular pressure was changed stepwise in the following sequence: 10, 9, 8, 7, 6, 5, 4, 5, 6, 7, 8, 9, 10, 12, 14, 16, 18, 20, 22, 20, 18, 16, 14, 12, and 10 mm. Hg. Allowance was made at each level for a steady flow to be obtained into or out of the perfusion reservoir. In the opposite eye, the perfusion was carried out similarly except that the sequence of pressure level settings was reversed. The flow from the perfusion reservoir was averaged for the two eyes at each intraocular pressure level. In a seprate group of 7 animals, angular venous pressure was measured under the same anesthetic system using a small polyethylene cannula.

Results

Table I is a summary of the average flow from the reservoir at each intraocular pressure level for each animal. The figure is a graph of the means and the standard errors for the entire group of animals. The least squares linear fit of the data points below angular venous pressure and the data points above angular venous pressure are also shown. A significantly higher slope was found at intraocular pressures greater than angular venous pressure where the mean slope was 0.66 ± 0.11 μL min.$^{-1}$ mm. Hg^{-1}. The slope of the pressure flow curve below angular venous pressure was found to be 0.30 ± 0.06 μL min.$^{-1}$ mm. Hg^{-1}.

Discussion

It has been shown that the intraocular pressure, P_i, is a function of the secretory rate of aqueous humor, F_s, the volume flux across the blood-aqueous barrier due to ultrafiltration and colloid reabsorption, F_j, the conventional facility to bulk outflow through the outflow channels, C_c, the uveoscleral flow into the suprachoroidal space, F_u, and the level of episcleral venous pressure, P_e. At steady-state, inflow into the eye equals outflow so that

$$F_s + F_j = (P_i - P_e)\, C_c + F_u \qquad (1)$$

If F_r is the flow into or out of the eye from a perfusion reservoir, then Equation 1 becomes

$$F_s + F_j + F_r = (P_i - P_e)\, C_c + F_u \qquad (2)$$

or

$$F_r = (P_i - P_e)\, C_c - F_s - F_j + F_u \qquad (3)$$

Bárány has shown that F_j is a function of intraocular pressure for P_i greater than P_e such that

$$\frac{\partial F_j}{\partial P_i} = C_p = \text{pseudofacility} \qquad (4)$$

Since P_e, F_s, and F_u are independent of intraocular pressure,* the first partial derivative of Equation 3 with respect to intraocular pressure is

$$\frac{\partial F_r}{\partial P_i} = \left(\frac{\partial P_i}{\partial P_i} - 0\right) C_c - 0 - \frac{\partial F_j}{\partial P_i} \qquad (5)$$

*F_u is apparently independent of intraocular pressure above 4 mm. Hg.[8]

Table I

	Intraocular pressure (mm. Hg.) (Flow, μL min.⁻¹)												
Monkey No.	*4*	*5*	*6*	*7*	*8*	*9*	*10*	*12*	*14*	*16*	*18*	*20*	*22*
1	-5.1	-3.4	-2.6	-2.2	-1.9	-1.5	-1.0	-0.3	0.5	1.6	2.8	3.9	6.3
2	-1.4	-1.1	-1.0	-0.9	-1.0	-0.2	0.5	2.3	4.7	6.6	8.3	9.7	11.0
3	-1.8	-1.7	-1.3	-1.2	-0.8	-0.5	-0.5	0.2	1.3	2.5	3.7	4.8	5.9
4	-2.7	-1.9	-1.8	-1.7	-1.3	-0.9	-0.9	1.0	2.5	3.8	4.3	5.6	5.7
5	-5.7	-3.8	-3.2	-2.5	-2.3	-1.9	-2.1	-1.4	-0.6	1.1	2.1	3.7	4.3
6	-2.4	-1.7	-0.7	-0.7	-0.6	-0.3	0.1	0.6	1.8	3.8	6.3	8.2	9.5
7	-3.1	-1.4	-0.8	-0.7	-0.6	-0.3	-0.3	0	0.8	2.1	3.7	5.4	6.9
8	-3.6	-2.4	-1.8	-1.6	-1.3	-1.1	-1.0	-0.9	-0.5	0.1	1.1	2.0	2.8
9	-1.8	-1.1	-0.6	-0.5	-0.1	0.4	0.9	2.8	4.4	5.6	6.9	8.0	8.9
Mean	-3.07	-2.06	-1.53	-1.33	-1.10	-0.70	-0.48	0.48	1.66	3.02	4.36	5.70	6.81
S.D.	1.50	0.97	0.90	0.71	0.69	0.72	0.91	1.39	1.92	2.12	2.37	2.48	2.59
S.E.	0.50	0.32	0.30	0.24	0.23	0.24	0.30	0.46	0.64	0.71	0.79	0.83	0.86

or

$$\frac{\partial F_r}{\partial P_i} = C_e + C_p = C_T. \qquad (6)$$

In more familiar terms

$$\frac{\Delta F_r}{\Delta P_i} = \text{conventional facility} + \text{pseudofacility} = \text{total facility} \qquad (7)$$

This relation is the basis for the determination of total facility by constant pressure or constant rate perfusion.

Equation 7 applies only for $P_i \geqq P_e$. If $P_i \leqq P_e$, the assumptions of the Bárány model are no longer valid. In order to extend the model to regions of intraocular pressure below episcleral venous pressure, we must look at F_j in detail.

The passive volume flux across a biologic membrane system can be estimated by the van't Hoff equation

$$F_j = C_j [\Delta P] - C_j \left[\sum_{i=1}^{n} \sigma_i RT (\Delta M_i) \right] \qquad (8)$$

where ΔP is the hydrostatic pressure difference across the membrane and where the expression

$$\sum_{i=1}^{n} \sigma_i RT (\Delta M_i)$$

represents the osmotic pressure difference across the membrane.[4-6] The first term represents "ultrafiltraton" and the second term represents "colloid reabsorption." Bárány has shown that the van't Hoff equation for the vascular tree of the eye can be written as

$$F_j = C_j [(P_a - P_v) X_c] - C_j [P_c]^{\circ} \qquad (9)$$

where the bracketed terms are the pressure difference terms and P_v is the terminal venous pressure as blood leaves the eye.

Suppose intraocular pressure equals episcleral venous pressure. Then $P_v = P_e$ and Equation 9 becomes

$$F_j = C_j [(P_a - P_e) X_c] - C_j [P_c] \qquad (10)$$

If the intraocular pressure now drops below P_e, a new term must be added to account for the additional ultrafiltration which will occur as a result of an increase in the hydrostatic gradient across the wall of the intraocular vascular tree. This term can be deduced from Equation 8 and is

$$C_j (P_e - P_i) \qquad (11)$$

Therefore Equation 10 becomes, $P_i < P_e$

$$F_j = C_j [(P_a - P_e) X_c] - C_j [P_c] + C_j (P_e - P_i) \qquad (12)$$

Since P_a, P_e, X_c, P_c, and C_j are independent of P_i

$$\frac{\partial F_j}{\partial P_i} = C_j, \; P_i < P_e \qquad (13)$$

[°]For an exact definition of P_a and X_c see Bárány.[1]

Table II

Animal, organ, method	*Filtration coefficient (μL min.$^{-1}$ mm. Hg^{-1} Gm. tissue^{-1})*	*Investigators*
Man, forearm (plethysmography)	0.057	Krogh, Landis, and Turner, 1932[15]
Dog, hind leg (perfusion)	0.061	Pappenheimer and Soto-Rivera, 1948[16]
Cat, hind leg (perfusion)	0.105	Pappenheimer, Renkin, and Borrero, 1951[10]
Dog, hind leg (perfusion)	0.058	Guyton, Prather, Scheel, and McGehee, 1966[17]
Rhesus monkey, eye (mannitol osmotic transient)	0.050°	Brubaker and Riley, 1972[2]
Rhesus monkey, eye (dextran osmotic transient)	0.228°	Brubaker and Riley, 1972[2]
Rhesus monkey, eye (perfusion)	0.075°	Present study

°Calculated on basis that weight of rhesus eye $\cong$ 4 Gm.

Moreover, since reverse flow ordinarily cannot occur through the conventional pathways, the term $(P_i - P_e)\ C_c$ becomes 0 in Equation 3, and we have

$$F_r = -F_s - F_j + F_u \qquad (14)$$

Differentiating Equation 14 with respect to P_i, we have

$$\frac{\partial F_r}{\partial P_i} = -0 - \frac{\partial F_j}{\partial P_i} + 0 = C_j \qquad (15)$$

or for finite changes

$$\frac{\Delta F_r}{\Delta P_i} = C_j,\ P_i < P_e. \qquad (16)$$

From the foregoing discussion, it can be seen that if the flow from an external reservoir, F_r, is plotted as a function of P_i, the slope of the curve is discontinuous at $P_i = P_e$. Above this point, the slope is equal to total facility, C_t, and below it, the slope is equal to the filtration coefficient, C_j.

From the least squares linear fit to the lower portion of the curve, the value of C_j is seen to be 0.30 ± 0.06 μL min.$^{-1}$ mm. Hg^{-1}. This value is in good agreement with the estimate obtained using osmotic transient experiments in the same animal.[2]

From the upper portion of the curve the value of C_t, total facility, can be calculated and is found to be 0.66 ± 0.11 μL min.$^{-1}$ mm. Hg^{-1}, a value in good agreement with measurements made in rhesus monkeys under deep barbiturate anesthesia[2, 3, 11] or in enucleated rhesus eyes,[12] but higher than values found under lighter anesthesia at higher perfusion pressures.[13]

The discontinuity in the slope of the flow-pressure curve appears to occur at a level higher than angular venous pressure. It has been shown that angular venous pressure very closely mimics episcleral venous pressure in some primate species,[3] but that the relationship may be variable in other primates.[7] The difference between angular venous pressure and the pressure at which the discontinuity occurs is probably not significant. If it were, the difference might be explained either by differences in episcleral and angular venous pressures or by a critical-closure phenomenon in the conventional outflow pathway for aqueous humor.

Perfusions were not carried out below 4 mm. Hg for two reasons. First, it is believed that uveoscleral flow is a function of intraocular pressure below 4 mm. Hg.[8] Second, the viscoelastic behavior of the globe at very low pressures introduces large uncertainties regarding the achievement of steady-state perfusion conditions.

The filtration coefficient of any membrane system is a measure of the solvent flux expected to occur across the membrane under hydrostatic and osmotic pressure gradients. The magnitude of the filtration coefficient found for the eye suggests that transient but physiologically sig-

nificant water flux occurs directly across the wall of intraocular blood vessels following changes in intraocular pressure or changes in blood-osmotic pressure, a supposition supported by clinical and experimental observations. These data are quantitatively compatible with the opinion that the negative feedback effect of modest changes in intraocular pressure on aqueous formation can be caused by a shift in net water flux across the blood vessel wall and need not be caused by an alteration in the rate of secretory transport systems. Moreover, the dramatic effect of systemically administered hypertonic or hypotonic agents on intraocular pressure and on intraocular tissue volume is indirect evidence that the filtration coefficient of the vessel system within the eye is physiologically important and that at least some portion of the vessel system is penetrated poorly by osmotic agents.

It must be pointed out that the measurements made in this study pertain to all of the vessels within the eye, and in no way demonstrate differences which would be expected to be found among anatomically different portions of the intraocular circulatory system. Bill[14] has measured the filtration coefficient of the iris vessels alone in vervet monkeys and reports a value of 0.0086 μL min.$^{-1}$ mm. Hg^{-1}, which is, as expected, a small percentage of that for the whole eye. Filtration coefficients for the isolated vessel systems of the ciliary body, the choroid, or the retina have not been reported. Comparison of the filtration coefficient of the whole eye to that of other vessel systems is shown in Table II. Filtration coefficients for isolated capillaries have been reported,[9] but are not directly comparable to the experimental data reported here.

REFERENCES

1. Bárány, E. H.: A mathematical formulation of intraocular pressure as dependent on secretion, ultrafiltration, bulk flow, and osmotic reabsorption of fluid, INVEST. OPHTHALMOL. **2**: 584, 1963.
2. Brubaker, R. F., and Riley, F. C.: The filtration coefficient of the blood-aqueous barrier, INVEST. OPHTHALMOL. **11**: 752, 1972.
3. Brubaker, R. F.: The measurement of pseudofacility and true facility by constant pressure perfusion in the normal rhesus monkey eye, INVEST. OPHTHALMOL. **9**: 42, 1970.
4. Van't Hoff: Une propriété générale de la matiere diluee, Su. Vet-ak-s Handlingor. **21**: 43, 1886.
5. Vargas, F., and Johnson, J. A.: Permeability of rabbit heart capillaries to nonelectrolytes, Am. J. Physiol. **213**: 87, 1967.
6. Durbin, R. P.: Osmotic flow of water across permeable cellulose membrane, J. Gen. Physiol. **44**: 315, 1969.
7. Bárány, E. H.: Topical epinephrine effects on true outflow resistance and pseudofacility in vervet monkeys studied by a new anterior chamber perfusion technique, INVEST. OPHTHALMOL. **7**: 88, 1968.
8. Bill, A.: Further studies on the influence of the intraocular pressure on aqueous humor dynamics in cynomolgus monkeys, INVEST. OPHTHALMOL. **6**: 364, 1967.
9. Landis, E. M., and Sage, L. E.: Fluid movement rates through walls of single capillaries exposed to hypertonic solutions, Am. J. Physiol. **221**: 520, 1971.
10. Pappenheimer, J. R., Renkin, E. M., and Borrero, L. M.: Filtration, diffusion, and molecular sieving through peripheral capillary membranes, Am. J. Physiol. **167**: 13, 1951.
11. Brubaker, R. F., and Kupfer, C.: Determination of pseudofacility in the eye of rhesus monkey, Arch. Ophthalmol. **75**: 693, 1966.
12. Peterson, W. S., Jocson, V. L., and Sears, M. L.: Resistance to outflow in the rhesus monkey eye, Am. J. Ophthalmol. **72**: 445, 1971.
13. Armaly, M. F.: Aqueous outflow facility in monkeys and the effect of topical corticoids, INVEST. OPHTHALMOL. **3**: 534, 1964.
14. Bill, A.: The role of the iris and the ciliary body in the drainage of aqueous humor, II. Studies in monkeys, Exp. Eye Res. **12**: 373, 1971. (Abst.)
15. Krogh, A., Landis, E. M., and Turner, A. H.: The movement of fluid through human capillary wall in relation to venous pressure and colloid osmotic pressure of the blood, J. Clin. Invest. **11**: 63, 1932.
16. Pappenheimer, J. R., and Soto-Rivera, A.: Effective osmotic pressure of the plasma proteins and other quantities associated with capillary circulation in the hindlimbs of cats and dogs, Am. J. Physiol. **152**: 471, 1948.

17. Guyton, A. C., Prather, J., Scheel, K., et al.: Interstitial fluid pressure. IV. Its effect on fluid movement through the capillary wall, Circ. Res. **19**: 1022, 1966.

DEFINITIONS

Pressure (millimeters of Hg)

P_a Mean arterial blood pressure measured near the origin of the ophthalmic artery.

P_i Intraocular pressure measured in the anterior chamber.

P_v Pressure in the intraocular veins measured at the point of exit of the vein from the eye.

P_e Pressure in the episcleral veins.

P_c Colloid osmotic pressure across the blood-tissue barrier (oncotic pressure).

Flow (μL min.$^{-1}$)

F_s Secretory rate of aqueous humor (independent of P_i).

F_c Conventional bulk outflow of aqueous humor through the anterior chamber angle.

F_J Volume flux across the blood-tissue barrier due to ultrafiltration-colloid reabsorption.

F_r Flow from external source into anterior chamber through a needle (perfusion flow).

F_u Uveoscleral flow of Bill,[8] bulk flow of fluid from the anterior chamber into the suprachoroidal space and through the sclera.

Conductance (μL min.$^{-1}$ mm. Hg^{-1})

C_c Conventional facility, or conductance of fluid to hydrostatic pressure differences across the conventional outflow channels at the anterior chamber angle ($C_c = \partial F_c/\partial P_i$).

C_p "Pseudo" facility, or conductance of fluid to hydrostatic pressure differences across the blood-tissue barrier ($C_p = \partial F_J/\partial P_i$).

C_t Total facility, $C_c + C_p$.

C_J Filtration coefficient for volume flux across the blood-tissue barrier under a hydrostatic or oncotic pressure gradient.

Miscellaneous

X_c Pressure index of "equivalent" capillary,* no dimensions ($X_c = C_p/C_J$).

σ = Staverman reflection coefficient, no dimensions ($0 \leqq \sigma \leqq 1$).

R = gas constant, μL mm. Hg μmole^{-1} degree^{-1}, = 62.36

T = absolute temperature, °K.

M = concentration, μmoles μL^{-1}.

Assumptions

$P_v = P_i$ for all $P_i \geqq P_e$ (1)

$P_v = P_e$ for all $P_i < P_e$ (2)

$F_c = 0$ for $P_i \leqq P_e$ (3)

$\partial P_e/\partial P_i = 0$ (4)

$\partial P_a/\partial P_i = 0$ (5)

$\partial C_c/\partial P_i = 0$ (6)

$\partial F_s/\partial P_i = 0$ (7)

$\partial F_u/\partial P_i = 0$ for all $P_i > 4$ mm. Hg (8)

*For exact definition of X_c, see Reference 1.