On the Mechanism for Flow in the Efferent Ducts

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A cilio-peristaltic model for pumping of efferent duct (ED) contents that predicts the observed volumetric flow rates and is consistent with a positive pressure gradient toward the epididymis is proposed. The model predicts that the major contributor to fluid flow is the peristaltic component. The cilia, in contrast, may reduce flow by creating reflux. This effect may be reduced considerably with an increase in fluid viscosity, which is in turn a function of sperm concentration. It is concluded from this investigation that if the peristaltic wave has a 33% or more constriction, the spermatozoan concentration in the ED is at least 4×10^8 cells cm^{-3} and flow rates in the ED are the same as in the rete testis; then transit in the ED is due to cilio-peristaltic pumping.

Key words: efferent ducts, peristalsis, cilia, fluid flow, mathematical model.

The efferent ducts (ED) are the only region of the male reproductive tract offering three completely separate propulsive mechanisms: smooth muscle, cilia, and secretion. In addition, their narrow diameters and multiplicity make these tubules major candidates for control of flow from the testis to the caput epididymis. The ED cilia have been suggested as a prime mover of tubal fluid since at least 1857 (Becker, 1857, as cited in Lucas, 1932). Setchell (1970) has concurred with this view, but Hargrove et al (1977) and Ellis et al (1978) present circumstantial evidence that the contractions of the testicular lamina propria and secretion are more reasonable propulsive force generators for the movement of sperm suspensions from the testis to the caput epididymis. As long as no direct measurements of ED motility and flow are available, the relative contributions of each pumping mechanism to tubal fluid flow can only be estimated. To the degree that our estimates conform both to relevant available data and the principles of fluid mechanics, however, we can be confident that our course of investigation is valid. Propulsion of ED fluid by cilia has been modeled (Lardner and Shack, 1972; Blake, 1973), and the predicted volumetric flow rates (Q) compare well with measurements from catheterized rete testes (Tuck et al, 1970). Unfortunately, it appears there are no measurements of flow generated by ciliated epithelium to assure the physiologist that the theoretical Q values were not merely the result of manipulating the parameters of the model.

In the present report we use more recent data from studies of the male reproductive tract, relevant measurements from studies of ciliated epithelium, and applicable fluid mechanical and rheological models to assess the potential roles of peristalsis and ciliary pumping in ED bulk flow generation.

Materials and Methods

Theoretical Models

Three theoretical models are used to predict Q in an ED:

1) The ciliary pumping model of Winet and Blake (1980) as applied to tubes (via the technique of Blake, 1973).

$$Q = \frac{\pi D'^2 U_m}{4} - \frac{\pi D'^4 \nabla P}{128\eta} \qquad (1)$$

where U_m is the maximum fluid particle velocity, D' the tube lumen diameter outside the cilia sublayer, η the

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suspension viscosity and ∇P the axial pressure gradient.

2) The peristaltic pumping model of Shen (1976)

$$\bar{Q} = \pi \bar{A} c \left[1 + \frac{Q^2}{2} - \frac{M_2}{M_4} - \frac{k_o \bar{R}_e}{8 M_4} \right] \quad (2)$$

where A is the average cross-sectional area of the relaxed tube, c the peristaltic wave velocity, $\phi = (1 - d)/D'$ the occlusion factor with d the tube diameter at deepest constriction, $k_o = (\nabla P/T)$ with ∇P the pressure difference over one wavelength, and $T = \lambda c^{-1}$ the wave period, $\bar{R}_e = (c \bar{D}^2 \rho / 4\eta\lambda)$ with $\bar{D}$ the relaxed ED diameter, λ the peristaltic wavelength, η the coefficient of viscosity and ρ the fluid density; $M_2 = (1 - \phi^2)^{-1.5}$ and $M_4 = (1 + 1.5\phi^2)(1 - \phi^2)^{-3.5}$.

3) The suspension viscosity model of Lightfoot (1974)

$$\eta = \eta_o (\Lambda \delta + 1) \quad (3)$$

where η_o is the viscous coefficient of the suspending medium, δ is the sperm volume fraction (the true spermatocrit) = VC with V the volume of one spermotozoan, C the sperm concentration, and Λ a crowding factor based upon random orientation and the length-to-width ratio a/b of the spermatozoan.

Experimental Models

The data used to quantitatively evaluate ED pumping are obtained from previous studies of the ED and/or other systems in the rat and frog ciliated epithelium. Data from ciliated epithelium of non-rat systems are considered appropriate for this study because of the similarity of this tissue in a variety of organisms, as shown in Table 1. We recognize that this simplifying assumption is a large approximation, but comparative studies support the notion that such characteristics as beat and beat frequencies are quite similar for all metozoa (Sleigh, 1974). The accessory tube geometry used for the study is presented in Fig. 1, and the constant values of relevant quantities are: $a = 7.0 \times 10^{-3}$ cm (Austin, 1965); $b = 2.3 \times 10^{-4}$ cm (Brennen and Winet, 1977); $c = 1.5 \times 10^{-1}$ cm sec^{-1} (Rukebusch and Fioramonti, 1975); $\bar{D} = 5 \times 10^{-3}$ cm (calculated from Reid and Cleland, 1957); $U_m = 3.75 \times 10^{-3}$ cm sec^{-3} (Winet and Blake, 1980).

In addition, the following approximations are assumed: $\rho = 1.0$ g cm^{-3}, $\lambda = 1.0$ cm, and, where not varied, $\eta = 1 \times 10^{-2}$ poise. Also, it is assumed that the ciliary effective stroke is epididymidad. This assumption is more a convenience than a critical condition for the model to be developed.

Pumping is evaluated by comparison of the published volumetric flow rates in the rete testis with $\bar{Q}$ generated at various pressure gradients as functions of ϕ and/or η.

As references for evaluating theoretical prediction of rat ED pumping, we use the rete $\bar{Q}$ value range calculated per tubule $3.0 \times 10^{-7} \leq \bar{Q} \leq 1.67 \times 10^{-6}$ cm^3 sec^{-1} (Free and Jaffe, 1979; Tuck et al, 1970) and the ∇P value range $2.3 \times 10^3 \leq \nabla P \leq 1.9 \times 10^4$ dyne cm^{-3} obtained from the rat rete measurements of Free and Jaffee (1979) and hamster and guinea pig caput measurements of Johnson and Howards (1975, 1976). (We assume here that the measurements from hamster and guinea pig are as close to the parallel values in the rat as they are to each other and that the ED length is 0.3 cm (Setchell, 1970).)

Results

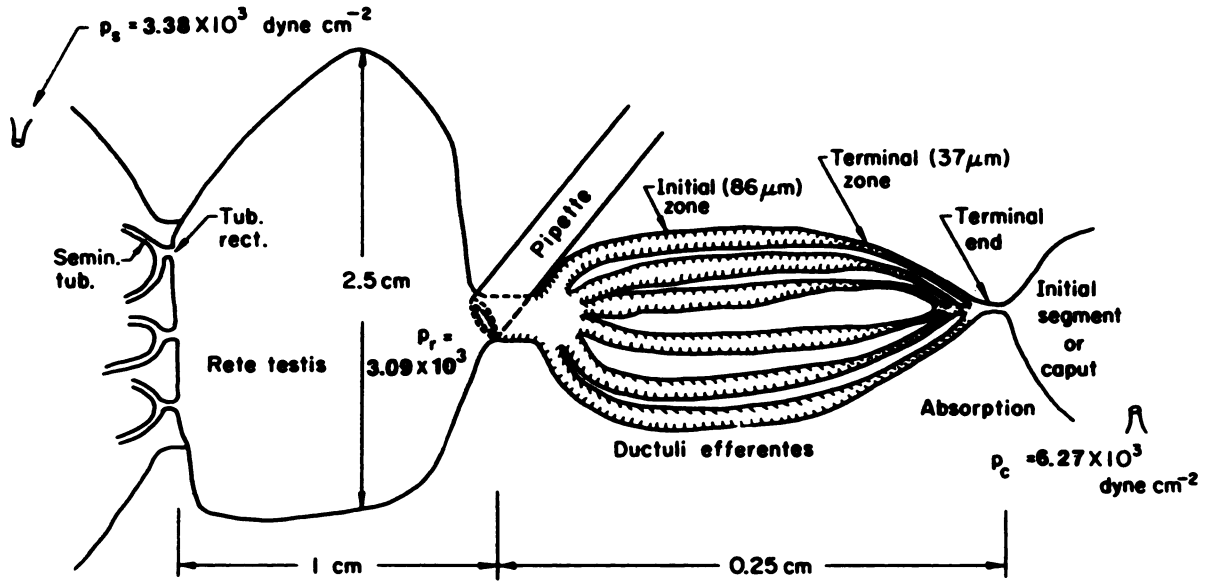
Ciliary Pumping

Figure 2 shows the relationship between Q and ∇P calculated from (1) for a variety of bulk viscosity values. The "measured" ∇P range is marked off with a bar at the top of the figure, and it will be noted that Q is negative (ie, flow is toward the rete) for the ∇P values of interest. If the viscosity of the sperm suspension is greater than 10^{-1} poise, however, the value of Q is positive but η must increase beyond 8.5×10^{-1} poise to produce a Q value near the midpoint of the range of measured Q values for the rat (indicated by the vertical bar at the right).

TABLE 1. Frog Ciliated Epithelium as a Model for Mammalian Ciliated Epithelium

Animal Organ	Cilium Length (μm)	Cilia Packing (cilia μm^{-2})	Metachrony	Beat Frequency (Hz)
Frog palate model	5–7*	3–11*	antiplectic	7–20
Oviduct				
Mouse/rabbit	5–7	8	antiplectic	20
Rat lung	5	6–10	?	22
Efferent ducts				
Hamster	6.8†	3–11†	?	?
Rat	5.5	?	?	?
Macaque	8.3‡	4.3–5§	?	?

References: * Brennen and Winet (1977); † Flickinger et al (1978); ‡ Hamilton (1975), § Ramos and Dym (1977).



INTERMEDIATE ACCESSORY TUBULES OF RAT
(not to scale)

Fig. 1. Schematic of male rat intermediate accessory tubes. Pressure values P_s (seminiferous tubules) and P_c (caput epididymis) are averaged from measurements from the hamster and guinea pig (Johnson and Howards, 1975, 1976), and P_r (rete testis) value is from the rat (Free and Jaffe, 1979). Other measurements are from the rat (Reid and Cleland, 1957; Setchell, 1970). The pipette is included to illustrate the region from which reported Q measurements are obtained.

Bulk Viscosity

The non-sperm components of rete fluid (see Setchell, 1970) are too soluble or dilute to significantly alter bulk viscosity. Spermatozoa, in contrast, act as long, slender particles that interact with one another to alter suspension bulk viscosity. The bulk viscosity of a suspension of particles is determined primarily by four factors: 1) shape and size of particles; 2) orientation of the particles; 3) interaction of the particles as indicated by mean free path; 4) flexibility of the particles. We assume that the spermatozoa are rigid and randomly oriented to facilitate calculations so that all one needs to obtain a working η reference value is cell-effective geometry (its viscous shape) and spermatocrit. We can obtain the essentials of the geometry by modeling rat sperm as prolate spheroids (needles), as shown in Fig. 3, in order to obtain the values of a and b needed to calculate Λ (Lightfoot, 1974) of (3). The volume of the sperm is

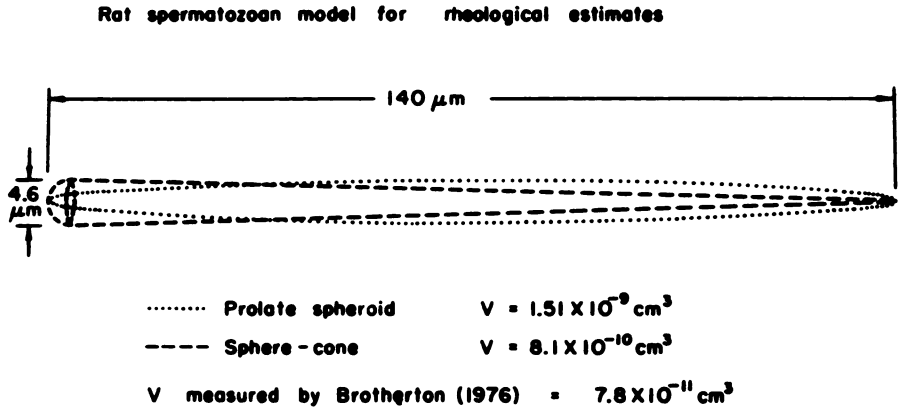


Fig. 2. Three models for the shape of a nonmotile rat spermatozoon in suspension. The prolate spheroid takes into account the viscous nature of the moving suspension. The sphere-cone (ie, semi-sphere cone) approximates the solid shape. The volume (V) measured by Brotherton (1976) is presented for comparison.

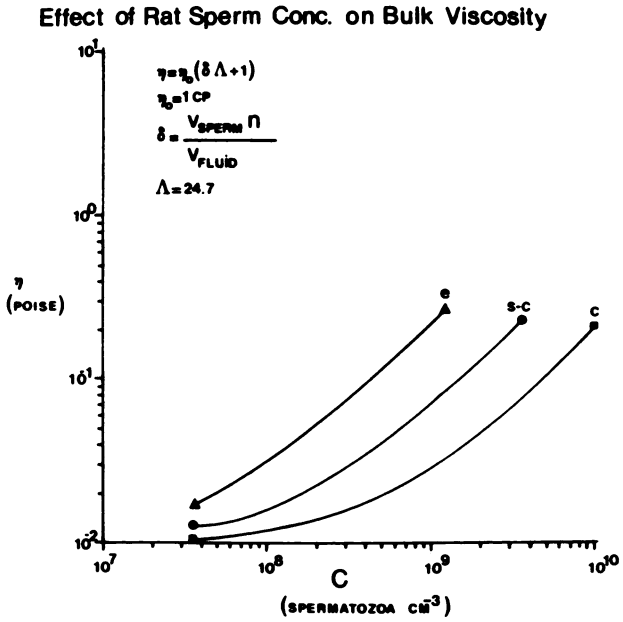


Fig. 3. The effect of the models of a rat spermatozoon presented in Fig. 2 on suspension viscosity (bulk) as a function of concentration. Symbols: e = prolate spheroid (ellipsoid), s-c = semi-sphere cone, c = Coulter counter volume measurement (Brotherton, 1976).

then required to calculate δ, and this value is obtained by 1) using the prolate spheroid model, which gives a "body" 1.4×10^{-2} cm long and 4.6×10^{-3} cm wide. Its volume is probably closest to the effective viscous volume (see Discussion section for more detail); 2) modeling a rat spermatozoan as a 4.6×10^{-3} cm wide semispherical head attached to a 1.4×10^{-2} cm long circular cone with a 4.6×10^{-3} cm wide base; 3) the method of Brotherton (1976), by use of a Coulter counter.

The change in bulk viscosity calculated by (3), as a function of sperm concentration C for each of these volumes, is plotted in Fig. 4. Depending on the volume model used, a minimum C of from 4.2×10^8 to 4.8×10^9 cells cm^{-3} would be required to create a bulk viscosity of 10^{-1} poise. There are, apparently, no measurements of C in ED, but reported values for the rat rete range 3.4×10^7 (Tuck et al, 1970) $\leq C \leq 1 \times 10^8$ cells cm^{-3} (Burgos and Tovar, 1974) and for the caput 1.4×10^8 (Gupta et al, 1974) $\leq C \leq 6.6 \times 10^8$ cells cm^{-3} (Turner and Howards, 1977). Burgos and Tovar (1974) reported a C value of 3.6×10^9 cells cm^{-3} for the rat "epididymis" without specifying the site.

Peristaltic Pumping

Theoretical values of $\bar{Q}$ generated by peristaltic pumping as described in (2) for various occlusion factors ϕ are plotted in Fig. 5. The effects of ∇P and η are so small in the ranges $0.01 \leq \eta \leq 0.85$ poise and $10^1 \leq \nabla P \leq 10^5$ dyne cm^{-3} that $\bar{Q}$ is constant at each ϕ. The effect of ϕ is significant but must exceed 0.10 to account for the higher $\bar{Q}$ values reported.

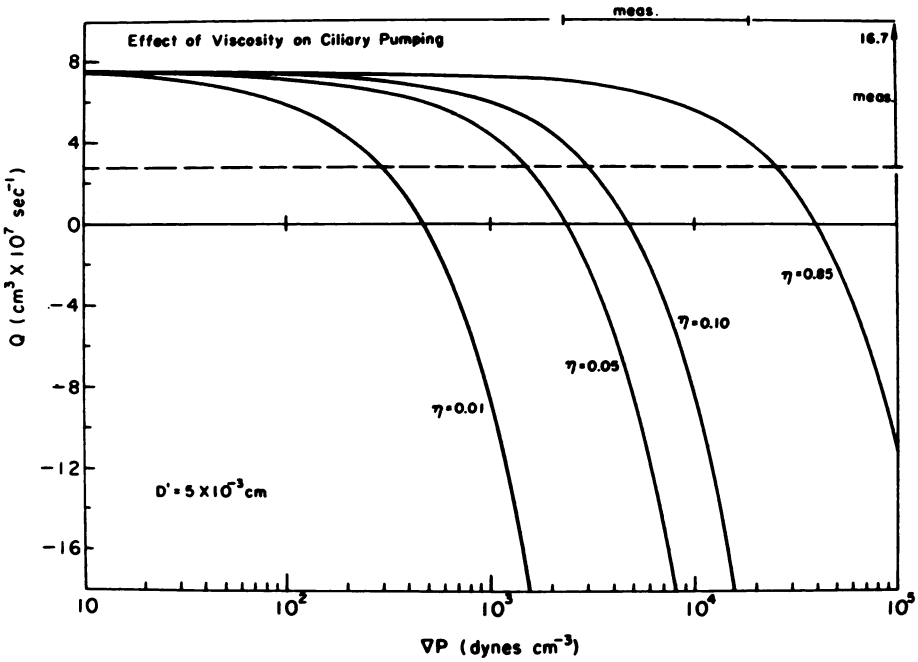


Fig. 4. Effect of viscosity on volumetric flow due to ED cilia alone in a single tubule as a function of ∇P. Negative values of Q indicate reflux toward the rete testis. The arrow at the right indicates the upper limit of the range of measured Q values $2.78 \times 10^{-7} \leq Q \leq 1.67 \times 10^{-6}$ cm^3 sec^{-1}.

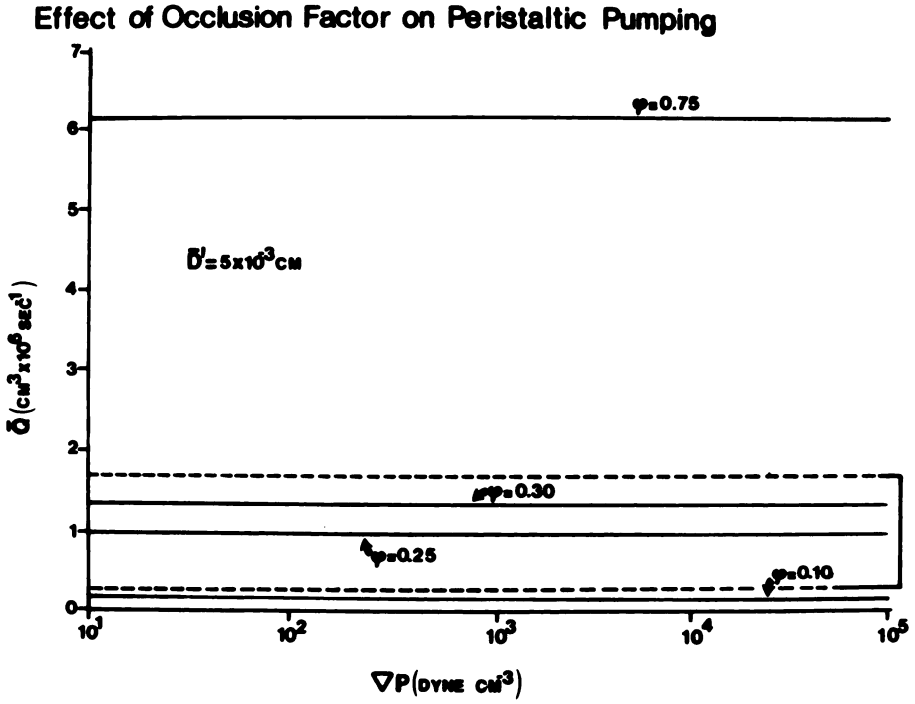


Fig. 5. The effect of occlusion factor (φ) on volumetric flow in a single ED tubule with peristaltic pumping only as a function of ∇P. The average volumetric flow rate $\bar{Q}$ has no negative values for this tube width. Also, viscosity and pressure gradient have no effect on $\bar{Q}$ over the ranges used in the present study. The vertical bar at the right indicates the range of measured Q values from the rat rete testis.

Combined Pumping By Cilia and Smooth Muscle

Theoretical values of Q for an ED with simultaneously pumping cilia and circular smooth muscle are presented in Fig. 6. The effect of ϕ on Q at the lower ∇P's is not influenced by η, but the opposite is true as ∇P increases. At higher ∇P values, viscous effects tend to dominate and Q values for all ϕ's tend to converge to a common value for a given η. In addition, reflux appears in tubes with ∇P values above 10^3 dyne cm^{-3} except where ϕ and ∇P are both highest. A summary of the effect of each pumping parameter on Q is presented in Table 2.

Conclusions

Given a waterlike viscosity for ductal fluid, there is no basis for attributing flow in the ED to ciliary pumping (see Fig. 2). By the same token, an occlusion of less than 20% would disqualify peristaltic pumping as the prime flow generator (see Fig. 3). The combination cilio-peristaltic pumping system shows a marked reduction in pumping ability at the lower η values because of the reflux generated by the cilia. This reflux effect is reduced by increasing η and/or ϕ, with the latter quantity having the greater effect. No observations of ϕ have been made for any of the male tubes, but the measurements cited above allow an estimate of C (the spermatozoan concentration in the ED). These measurements come from the rete testis and epididymis and, if we assume that C lies between the limits given, a conservative average for the ED from these data would be 4×10^8 spermatozoa cm^{-3}. Using a viscous boundary spermatozoan geometry, this yields a calculated $\eta \sim 10^{-1}$ poise (see Fig. 4). This viscosity is too small to allow ciliary pumping to move fluid through the ED at the faster rete Q rates (see Fig. 3). It does, however, reduce reflux sufficiently to allow a ϕ value of 0.33 to account for the entire range of observed Q values in the cilio-peristaltic system (see Figs. 5, 6).

Accordingly, we may conclude that if the peristaltic wave has a 33% or more constriction, the spermatozoan concentration in the ED is at least 4×10^8 cells cm^{-3} and that flow rates in the ED are the same as in the rete testis; transit in the ED is then due to cilio-peristaltic pumping.

Discussion

Convective flow in biological tubes is generated by three mechanisms: 1) pulsatile upstream pumping; 2) peristaltic tube pumping; 3) ciliary pumping.

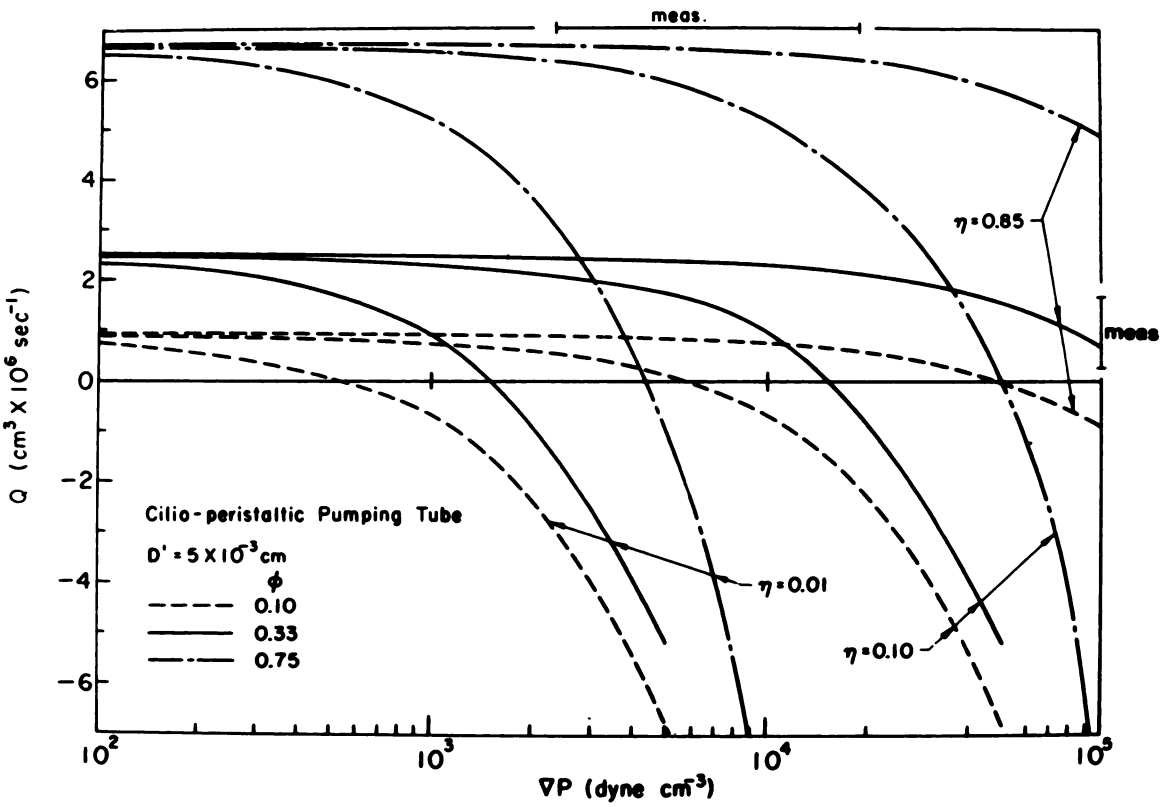


Fig. 6. The combined effect of cilia and peristalsis on Q in a single ED tubule for various ϕ and η values as a function of ∇P. At low ∇P values, ϕ dominates Q, but at high ∇P values η dominates Q. Measured ranges of Q (from the rete testis) and ∇P (calculated from P values presented in Fig. 1) are indicated by vertical and horizontal "error" bars, respectively. Note that a $\phi = 0.33$ and a $\eta = 0.1$ come quite close to generating the range of measured Q values within the range of "measured" ∇P values (for an ED length of 3.5 mm the values would fit).

Only in the first system is the upstream pressure greater than the downstream pressure. Since the opposite is true in all male accessory tracts measured thus far (Johnson and Howards, 1975, 1976), the least likely mechanism for ED flow is a prerete upstream pump such as the tunica albuginea or the lamina propria of the testis (Hargrove et al, 1977). This is not to say that the ∇P of the ED cannot be generated by a testis pump, but the ED flow cannot be maintained by such a pump against a positive ∇P. The rete testis is itself an unlikely candidate for flow generation, as it has too little smooth muscle. It would be possible to construct a model for flow generation by an absorption "pump" in the ED, initial segment, or caput; but again, the pressure gradient would have to be the reverse of the one measured.

TABLE 2. Effect on Q of Various Parameter Changes in Cilio-Peristaltic Tubules

Parameter	c	C	$\bar{D}$	∇P	Re	U_m	η	λ	ρ	ϕ	
Increase	+	+	−	−	−	+	+	+	−	+	Q
Decrease	−	−	+	+	+	−	−	−	+	−	Q

Since the ED contains both kinocilia and circular smooth muscle, it would be specious to ignore either in any pumping model. Similarly, the suspension moved by the pumps contains a high concentration of long, slender bodies that must affect the rheology of the tube. The present model has attempted to incorporate these three components to formulate some testable predictions, assuming the Q in the rete is the Q in the ED system. First, cilia can generate only the lowest measured Q values and require reasonable but high viscosities for this. Second, peristalsis can generate the necessary Q with an appropriate ϕ at any viscosity, but these pumps must overcome the reflux generated by cilia at the lower η. Accordingly, cilia may be viewed as actually decreasing Q at low η while aiding peristalsis at high η in the cilio-peristaltic system. Thus, with the addition of absorption, we have the basic elements of a control system. Let us suppose an occlusion factor range $0.25 \leq \phi \leq 0.35$

and a rat spermatozoan concentration of 1×10^8 cells cm^{-3} with all other parameters remaining as reported above. An examination of Figs. 4 and 6 will show that this system cannot move sperm into the epididymis. With the aid of an absorption of 90% (Crabo, 1965), the C value would rise to 1×10^9 cells cm^{-3}, which would create a η sufficient to reduce reflux to a value that would allow peristalsis to generate the necessary Q. Such a positive feedback control system may be viewed as a hydromechanical mechanism for maintaining high spermatocrits in the epididymis.

This model predicts other beneficial effects consistent with the suggestions of Turner and Howards (1977). The positive pressure gradient associated with cilio-peristaltic pumping enhances transmural transport in the same manner that transcapillary transport operates at the arteriole side of the vascular bed. Solute particles may move across membranes by diffusive (passive or active transport) or convective (solvent drag) mechanisms, but transport is faster by the latter process. Accordingly, a sufficiently large driving pressure (hydrostatic minus osmotic (Hobbie, 1978)) could aid absorption at the ED–caput junction significantly, or simply prevent solvent influx. Such transport, if selective, could explain the hyperosmolality observed by Johnson and Howards (1977) in the epididymis.

Finally, we must explain the predicted rheology of the spermatozoan suspension. Any solid in suspension carries a fluid coat that remains attached even if the solid moves at a speed unlike that of the suspending medium. This coat or envelope is the nonslip layer and acts effectively as an extension of the solid. The more viscous the fluid, the thicker the envelope. When the Reynolds number (the ratio of inertial to viscous stresses) is as small as its value for the present report, the envelope forms a spheroidal "viscous" shape that bears little resemblance to the solid it surrounds (Happel and Brenner, 1965). The "viscous" shape has a "viscous volume" that is a more valid measure for calculating Λ than either the semisphere-plus-cone model (which is probably closest to the true solid volume) or the volume measured by Brotherton (1976) by use of a Coulter counter (using a Coulter counter to estimate the volume of any particle that deviates from a sphere as much as a spermatozoan does is at least dubious). Accordingly, the curve to the far left (e) of Fig. 4 is used for calculating Λ and thereby, η. A more significant effect on η may be expected from an uneven radial distribution of spermatozoa in the ED, as suggested by the observations of Orgebin-Crist (1965). She found that caput spermatozoa in the rat are more concentrated near the tube wall, forming a dense ring or inner "flow tube." It is logical to assume that this radial distribution took place in the terminal ED. As a result, η would increase in this peripheral region, and the transit of spermatozoa could be enhanced to a point at which peristalsis would be unnecessary. By the same token, reflux in the center of the ED would continue and, indeed, would also be enhanced. The resulting forward-moving tube of spermatozoa adjacent to a core of refluxing spermatozoa could favor the distribution observed, that is, new sperm to the inside, the old sperm to the outside.

The cilio-peristaltic model described herein is a working hypothesis that presents some testable predictions about flow in a tube lined with cilia and smooth muscle. The reason the relevant observations have not been made for the ED is not due to lack of interest but rather to the difficulty of the required manipulations. The theory provides a means of checking measurements against predictions derived from the laws of fluid mechanics. If any of our assumptions are significantly incorrect, these laws contain the mechanism for adjusting the model without violating the rules of physical analysis. The ability of a physics model to guide an experimentalist to relevant measurements and the ability of experimental results to guide the biophysicist to a more precise model are the foundations of the multidisciplinary approach to physiological problems.

References

Austin CR. Fertilization. Englewood Cliffs: Prentice-Hall, 1965.

Becker O. Ueber Flimmerepithelium und Flimmerepithelium im Geschlechtsapparate der Saugethiere und des Menschen. Moleshotts Untersuch 1857; 2:77.

Blake J. Flow in tubules due to ciliary activity. Bull Math Biol 1973; 35:513–523.

Brennen C, Winet H. Fluid mechanics of propulsion by cilia and flagella. Annu Rev Fluid Mech 1977; 9:339–398.

Brotherton J. Difference in size between spermatozoa from the cauda epididymis and the caput epididymis of the rat. J Reprod Fert 1976; 48:365–366.

Burgos MH, Tovar ES. Sperm motility in the rat epididymis. Fertil Steril 1974; 25:985–991.

Crabo B. Studies on the composition of epididymal content in bulls and boars. Acta Vet Scand [Suppl] 1965; 6:1–94.

Ellis LC, Buhrley LE Jr, Hargrove JL. Species differences in contractility of seminiferous tubules and tunica albuginea

as related to sperm transport through the testis. Arch Androl 1978; 1:139–146.

Flickinger CJ, Howards SS, English HF. Ultrastructural differences in efferent ducts and several regions of the epididymis of the hamster. Am J Anat 1978; 152:557–585.

Free MJ, Jaffe RA. Collection of rete testis fluid from rats without previous efferent duct litigation. Biol Reprod 1979; 20:629–728.

Gupta G, Rajalakshmi M, Prasad MRM, Moudgal NR. Alteration of epididymal function and its relation to maturation of spermatozoa. Andrologia 1974; 6:35–44.

Hamilton DW. Structure and function of the epithelium lining the ductuli efferentes, ductus epididymidis and ductus deferens in the rat. In: Hamilton DW, Greep RR (eds.) Handbook of physiology. Section 7. Washington, DC: American Physiology Society, 1975:259–301.

Happel J, Brenner H. Low Reynolds number hydrodynamics. Englewood Cliffs: Prentice-Hall, 1965.

Hargrove JL, MacIndoe JH, Ellis LC. Testicular contractile cells and sperm transport. Fertil Steril 1977; 28:1146–1157.

Hobbie RK. Intermediate physics for medicine and biology. New York: John Wiley and Sons, 1978.

Johnson AL, Howards SS. Intratubular pressure in the testis and epididymis before and after vasectomy. Am J Physiol 1975; 228:556–564.

Johnson AL, Howards SS. Intratubular hydrostatic pressure in testis and epididymis before and after long-term vasectomy in the Guinea pig. Biol Reprod 1976; 14:371–376.

Johnson AL, Howards SS. Hyperosmolality in intraluminal fluids from hamster testis and epididymis: a micropuncture study. Science 1977; 195:492–493.

Lardner TJ, Shack WJ. Cilia transport. Bull Math Biophys 1972; 34:325–335.

Lightfoot EN. Transport phenomena and living systems. New York: John Wiley and Sons, 1974.

Lucas AM. Ciliated epithelium. In: Cowdry EV, (ed.) Special cytology. Vol. 1. New York: Hafner, 1932:407–474.

Orgebin-Crist MC. Passage of spermatozoa labelled with thymadine-^{3}H through the ductus epididymis of the rabbit. J Reprod Fertil 1965; 10:241–251.

Ramos AS Jr, Dym M. Ultrastructure of the ductuli efferentes in monkeys. Biol Reprod 1977; 17:339–349.

Reid BL, Cleland KW. The structure and function of the epididymis. I. The histology of the rat epididymis. Aust J Zool 1957; 5:223–246.

Rukebusch M, Fioramonti J. Electrical spiking activity and propulsion in small intestine in fed and fasted rats. Gastroenterology 1975; 68:1500–1508.

Setchell BP. Testicular blood supply, lymphatic drainage, and secretion of fluid. In: Johnson AD, Gomes WR, Vandemark NL, (eds.) The testis. New York: Academic Press, 1970:101–239.

Shen MC. Asymptotic theory for peristaltic transport in a tube of arbitrary cross section Phys Fluids 1976; 19:213–218.

Sleigh MA. Patterns of movement of cilia and flagella. In: Sleigh MA, ed. Cilia and flagella. New York: Academic Press, 1974:79–92.

Tuck RR, Setchell BP, Waites GMH, Young JA. The composition of fluid collected by micropuncture and cathaterization from the seminiferous tubules and rete testis of rats. Pfluegers Arch 1970; 318:225–243.

Turner TT, Howards SS. Sperm maturation, transport and capacitation In: Cockett ATK, Urry RK eds. Male infertility: workup, treatment and research. New York: Grune and Stratton, 1977: 29–57.

Winet H, Blake JR. On the mechanics of mucociliary flows. I. Observations of a channel model. Bioheology 1980; 17:135–150.