Quantitative Studies of Microcirculatory Structure and Function

II. DIRECT MEASUREMENT OF CAPILLARY PRESSURE IN SPLANCHNIC MESENTERIC VESSELS

By Benjamin W. Zweifach

ABSTRACT

A systematic study was made of capillary pressures in the microvascular bed of the mesentery of the cat. Simultaneous recordings were made with two micropipettes using a modification of the Wiederhielm servo-null system. All of the measurements were then referred to their specific location in the capillary bed by photographically reconstructing discrete networks. Pressures in different capillary branches within a given network ranged from 40 mm Hg to as low as 24 mm Hg. There was no obvious pattern in the distribution of high-pressure vessels relative to low-pressure vessels. When capillary pressure was measured in a selected vessel and averaged over a period of several minutes, it remained constant (±3 mm Hg), despite much greater fluctuations in the precapillary and postcapillary vessels. A number of different procedures was used to obtain average values for capillary pressure in a given network. Estimates of capillary pressure from venous wedge pressures in the mesentery were related to the actual capillary pressure, but the precise proportionality of the two pressures was not the same in different preparations. Possibly, the estimates of average capillary pressure based on direct measurements were higher than those obtained indirectly from calculations of fluid exchange because other factors affecting the latter were not adequately considered.

KEY WORDS microcirculation micropressures cat autoregulation mesenteric circulation wedge pressure micropipettes

Capillary pressure in mammalian tissues has, for the most part, been estimated indirectly from related hemodynamic phenomena. Several studies have been made on single capillaries (1, 2) but these studies have added little to the classical work of Landis (3), which dealt extensively with micropuncture of vessels in the frog mesentery and included only a comparatively small number of recordings from the mesentery of the guinea pig. The Landis method for recording mean pressure is crude compared with the fidelity of present-day electronic servo procedures. Wiederhielm (2) used an electronic micropressure technique to make single determinations in small vessels of the frog and in the wing of the bat (4). On the whole, these measurements fell within the range of 20 mm Hg to 30 mm Hg that has been cited for mammalian capillaries. Estimates of average pressures for the network of vessels involved in fluid exchange include data for both capillaries and venules and reflect the interaction of several variables whose contributions are not clearly spelled out. The average capillary pressure in skeletal muscle calculated by isogravimetric (5) or isovolumetric manipulations (6) ranges from 15 mm Hg to 20 mm Hg and is lower than that obtained in the cremaster muscle of the rat using the direct micropuncture procedure (7).

An analysis of basic hemodynamic factors in the microcirculation requires more precise and more extensive data on pressure relationships within the capillary network. In the present study, topographical maps of micropressure were prepared for the cat mesentery from direct recordings. Time-dependent changes were ascertained, and an attempt was made to determine the significance of averaged values of capillary pressure for the bed as a whole. Capillary pressures in different portions of a particular network differed by as much as 10–11 mm Hg, but the inhomogeneity of the pressure distribution made it difficult to apply such numeri-
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cal values to functional changes without additional information concerning the anatomical features of the network.

Methods

Measurements of capillary pressure ($P_c$) were made in the mesentery of the cat. Over 100 cats were studied, and as many as 20-30 separate readings were made in each preparation. Details of the experimental procedure have been described previously (8). The cats were anesthetized with sodium pentobarbital (30 mg/kg, iv). The splanchnic mesentery of the cat was maintained under controlled conditions for examination by intravital microscopy (9). Pressures were recorded by an electronic servo-micropipette system modified from that originally used by Wiederhielm et al. (10).

Direct measurement of $P_c$ is possible, but because of the small size of capillaries in the mesentery (5.8-8μ wide) the insertion of a micropipette, even one with a 1.0μ tip opening, interferes with blood flow and can give atypical readings. Therefore, most of the values of $P_c$ presented in this study were obtained by inserting the micropipette into a branching point so as to occlude the branch and form a static tube in which the end pressure reflected that in the unobstructed vessel. Some of these measurements were made with separate probes in two successive branches so that the pressure drop along the intervening length of the capillary could be recorded and values of $dP/dL$ could be calculated.

Results

TEMPORAL VARIATIONS IN CAPILLARY PRESSURE

A striking feature of the microcirculation is the intermittent nature of the blood flow; about 20-30% of the available capillary channels can have a markedly slowed flow or even remain static at any given moment. The periodicity in any given capillary is quite irregular; slowing and stoppage of red cell movements occur for anywhere from 5-10 seconds up to several minutes. In addition to these local changes, central arterial pressure occasionally varies as much as 15-20 mm Hg during 1-2-hour experiments. This degree of change in arterial pressure, however, had no significant effect on $P_c$. A comparison of the effect of individual differences in central pressure on the corresponding $P_c$ values showed no significant correlation between the two, as was the case for all other microvessels below 40μ in diameter (8).

The routine procedure in all of these experiments was to use a second micropipette in a feeding precapillary as a reference point for shifts in central driving pressure. When the pressures for a particular vessel were averaged at 20-second intervals over a period of 3-4 minutes, the values were quite reproducible. Thus the time-averaged pressures for 45 capillary vessels in cats with almost identical central pressures ($120 ± 10$ mm Hg) had a mean value of $32.3$ mm Hg with a standard deviation of only $3.2$ mm Hg. The widest fluctuations ($±4-7$ mm Hg) were found in the narrowest capillaries (5-6μ wide). Differences in $P_c$ of as much as 10 mm Hg were encountered in individual vessels within the same modular area and in different sectors of the mesentery. The vessels in which $P_c$ was comparatively high (40-45 mm Hg) maintained this level, as did those with low pressures (18-20 mm Hg), when averages over several minutes were obtained.

During observations of 30-40 minutes, two types of fluctuations were recorded. Short-lasting (15-20 seconds) sharp increases or decreases of 3-5 mm Hg were seen in $P_c$; there was no consistent pattern to these irregularities. In some cases steady state was interrupted by a substantial increase or decrease in $P_c$ (as much as 10-12 mm Hg) which developed gradually; this change took 5-8 minutes on the average to reach a peak and was then followed by a return to the steady-state level in 2-3 minutes. These substantial shifts in $P_c$ were heralded by a comparable change in the precapillary pressure. Dilation of the feeding vessels could also be detected by the sudden widening of the capillary pulse pressure. Pulse pressures in the true capillaries averaged 1-2 mm Hg. With arteriolar dilu-
gram for almost 400 direct measurements of $P_c$ in 73 cats. The somewhat skewed distribution was probably due to the inhomogeneity of the bed, the presence of shunt pathways, and the fact that capillary channels can empty into different venular tributaries, causing different pressure drops across the various capillaries. If the upper and lower limits shown in Figure 1 are assumed to reflect varying states of dilation or contraction in the bed as a whole, the boundary limits for pressure excursions in beds of this type under steady-state conditions fall between 22 mm Hg and 48 mm Hg. The calculated average $P_c$ in the mesentery of the cat was $31.7 \pm 6.4$ (SD) mm Hg.

In some preparations, intermittent precapillary vasomotion stopped capillary flow in the vessel’s dependent branches for 20-40 seconds. An attempt was therefore made in these cases to record concurrently the pressure in the capillary network and the venous draining vessel. In 26 such measurements, the pressure at the time flow stopped was reduced on the average by 2-5 mm Hg and frequently dropped to that of the downstream postcapillary. For example, in a given experiment $P_c$ averaged 31 mm Hg and, when flow stopped spontaneously, the pressure fell to 26 mm Hg.

TOPOGRAPHICAL DISTRIBUTION OF CAPILLARY PRESSURE

Direct measurements of $P_c$ were recorded with respect to type of vessel, diameter, and location of the vessel in the network as a whole. Figure 2 is a photomontage of a microvascular network which was examined in detail to determine pressure relationships within the capillary interconnections. Two of the vessels are shunts; they exhibited a more rapid flow and had a greater number of red blood cells. These vessels were somewhat wider (9-11 µ) than the remaining capillaries (5.5-7 µ).

In the larger vessels (arterioles, shunts, and venules), the micropressures were obtained by direct intubation. In the finer capillaries, micropipettes were inserted at branching points to record pressures. A reasonably complete mapping of pressures took 20-30 minutes to complete; therefore, the distribution pattern does not represent the pressures at any given moment. Out of approximately 35 vessels in the field, micropressures were obtained in 19.

The pressures in particular capillaries of the same network ranged from 41 mm Hg to 20 mm Hg (Fig. 2). Outflow pressures in the immediate post-capillaries were, however, more consistent (27-30 mm Hg). Capillary vessels, which arose directly from the arterioles, had pressures of 38-40 mm Hg; those distributed in the midregion averaged 29-30 mm Hg, and the most distal branchings showed $P_c$ values of 26-28 mm Hg. In this particular bed, the mean $P_c$ was $29.4 \pm 5.6$ mm Hg, and the most distal branchings showed $P_c$ values of 26-28 mm Hg.

SEQUENTIAL PRESSURES ACROSS THE CAPILLARY NETWORK

As part of a systematic analysis of the pressure distribution in different segments of the microcirculation, successive capillary branches were followed step by step from the arterial to the venous side. The accompanying photographic montage (Fig. 3) demonstrates the serial distribution of pressures expressed in cm H$_2$O. The capillaries in Figure 3 ranged from 7 µ to 9 µ wide. Red blood cells in the cat were measured by interference microscopy; they averaged about 6.0 ± 0.7 µ in diameter. The micropipettes were inserted into the junctional portion of the side branches of the vertically aligned vessel so as to occlude the branch. This capillary sequence contained four side branches, and the fall in pressure was about 2.0 cm H$_2$O from one nodal branching point to the other (usually about 250-300 µ).

Because a number of other parameters were available in the present study, it was useful to compare the observed pressure drop with that which could be calculated. In view of the comparatively slow movement of the blood and the passage of the red blood cells in single file along these narrow capillaries, it was comparatively simple to estimate average blood velocity from a video tape recording made when the micropressures were recorded with two microneedles. Four readings of red blood cell velocity were averaged and used to calculate flow; then the change in pressure was determined from the conventional Poiseuille formula for blood flow. The average velocity in these capillaries was $410 \pm 70$ µ/sec. Inasmuch as the viscosity of blood in the microcirculation is unknown, a viscosity slightly above that of plasma (2.0 centipoise) was assumed. The calculated reduction in pressure was 8-10 cm H$_2$O in contrast to the change in pressure of 15 cm H$_2$O that was actually measured. The approximately 70% difference between these values presumably is due to either the presence of side branches or the inaccuracy of estimates of the flow properties of the blood by a simple viscous term.

In addition to direct measurement of $P_c$, several other methods were used which did not involve...
Repeated puncture of the vessels. A particularly useful procedure was the mechanical obstruction of flow with a rounded needle either upstream or downstream from the recording micropipette (Fig. 4). For example, an occlusion that cut off the feeding arteriole left the bed open by way of the capillary network to the effluent venule and gave readings which reflected mean $P_c$. In Figure 4A, the flow in a 20$\mu$ arteriole was obstructed temporarily on the upstream side and pressure fell within 1–2 seconds from 40 cm H$_2$O to 26 cm H$_2$O, which is about 30% below the average capillary pressures measured directly in three vessels of this network. In Figure 4B, the converse type of obstruction was used. $P_c$ was recorded directly and the vessel was obstructed on the venous side.
leaving the network open only to the arteriolar circulation. In Figure 4C, the pressure was recorded in a postcapillary or collecting venule (24μ wide) that was obstructed on its downstream side, and $P_c$ was estimated at 31 cm H$_2$O.

**AVERAGE CAPILLARY PRESSURE**

A numerical average of $P_c$ based on the pressure drop across the capillary network could be obtained. In these instances, the arterial pressure was obtained by intubating a precapillary vessel (12–18μ) and the venous or postcapillary pressure was obtained by a pipette in the smallest collecting venule (15–20μ) (Fig. 5). The mean value for such average pressures was 33.8 ± 1 mm Hg. Such estimates tended to be higher than the directly measured pressures depending on how close to the precapillary inflow and the postcapillary outflow the actual recordings were made. The larger the intubated precapillary and postcapillary vessels, the greater the pressure drop between them and, hence, the greater the discrepancy with direct measurements.

The values obtained by the four methods used in the present study are summarized in Table 1. Depending on the accuracy which the experimental...
situation required, any one of the four methods employed gave reasonable estimates of mean capillary pressures. Obviously, the most informative data were obtained by direct sampling of a reasonable number of vessels. The significance of such averaged values to pressure-flow relationships or to fluid exchange was difficult to establish, especially in view of structural and possible functional differences between vessels in different portions of the capillary network.

VENOUS WEDGE PRESSURES AND CAPILLARY PRESSURE

In many tissues in which direct observation of the microcirculation was not possible, estimates of \( P_c \) were obtained by introducing a fine catheter into a peripheral vein and advancing it retrograde until

\[
\frac{1}{2}(P_A + P_V) = \text{Capillary pressure estimated by different methods.}
\]

### TABLE 1

<table>
<thead>
<tr>
<th>Procedure</th>
<th>No. of vessels</th>
<th>Cap mesentery (mm Hg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Topographical average for a complete network</td>
<td>19</td>
<td>29.4 ± 5.6</td>
</tr>
<tr>
<td>Numerical average in different animals</td>
<td>306</td>
<td>31.7 ± 6.4</td>
</tr>
<tr>
<td>Average of arteriolar and venular pressures</td>
<td>93</td>
<td>33.7 ± 6.2</td>
</tr>
<tr>
<td>Microobstruction</td>
<td>23</td>
<td>28.2 ± 5.7</td>
</tr>
</tbody>
</table>

All values are means ± SD.
Venous wedge pressures compared with direct measurement of pressure in the collecting venules (15–20μ) in the mesentery. The two pressures showed a similar trend, but the actual proportion of one to the other was difficult to establish a priori.

Discussion

Hydrostatic pressures in single blood capillaries have been determined by direct measurement in the frog mesentery (3), the guinea pig mesentery (11), the rabbit omentum (12), and the bat wing (4). Most of these measurements were repeated but not continuous, and there has been no concerted effort to obtain sufficiently precise samplings in vascular networks whose anatomical features have been worked out at the same time.

Average values for whole organs or masses of muscle tissue have been calculated as a reflection of the exchange of fluid between the blood and tissue compartments (14). Even in experiments on single capillaries with the Landis (3) microocclusion procedure, the pressures, which are calculated on the basis of the Starling constitutive equation, are not true hydraulic pressures but must be considered net or effective pressures because of the unknown contribution of the interstitial tissue to the exchange of fluid (13). In a similar context, isogravimetric or isovolumetric procedures (14) in perfused organs do not necessarily provide the hydrostatic Pe for skeletal muscle in vivo, since neither the hydrostatic nor the colloid osmotic forces outside the capillary are known.

As discussed previously (8), the fact that the micropressure measurements were made in the exteriorized mesentery lays open the possibility that vasodilation occurred and altered the true in vivo distribution of resistances. Evidence has been provided that these mesenteric preparations are functionally intact and unquestionably provide data well within the physiological range. Inasmuch as no other direct approaches are currently available which can provide equivalent in situ information under more ideal conditions, the present data at the very least serve as a useful beginning for the analysis of basic relationships within the microvasculature.

The pressure relationships in the splanchnic mesenteries are not necessarily the same as those in other parenchymal tissues. Microvascular pressures in the mesentery are elevated because of the peculiar circumstance created by drainage of the mesenteric circulation into the portal venous system. In the cat, portal venous pressures are 6–8 mm Hg as opposed to the 3–5-mm Hg pressures in the veins that drain other tissues of the body. In a previous study on the cremaster muscle of the rat (7), micropressures were 4–8 mm Hg below those in the mesentery of the cat.

The nonuniform distribution of pressures in different regions of the capillary network does not support the oversimplified schema adopted by most models whereby pressures on the arterial side are higher than the prevailing plasma colloid osmotic pressure and pressures on the venous side are brought below plasma colloid osmotic pressure by frictional losses so as to balance out fluid filtration. In the mesentery, the hydraulic pressure in over 80% of the capillaries with an active flow was above plasma colloid osmotic pressures until the venular effluent system was formed.

In line with the Starling formulation for transcapillary exchange of fluid, the two opposing forces, capillary hydrostatic pressure and plasma colloid osmotic pressure apparently are not in equilibrium in vessels which have an active flow. Rather than adopting the view that the pressures are skewed to the high side because of a dilation associated with exteriorization, the current findings might reflect that one role of the mesenteric...
Circulation is to provide fluid for the abdominal cavity. Capillary pressures fell below plasma oncotic pressures at irregular intervals primarily in vessels in which active flow was stopped for varying periods as a result of spontaneous narrowing of the precapillaries.

It has been proposed previously (8) that arteriolar-precapillary branchings, through a combination of physical factors and vasomotor adjustments, are the principal sites within the microcirculation at which pressure is modulated and brought within a comparatively fixed range. The pressure downstream within the capillary network then falls in accord with physical factors alone. Shifts in the pressure relationships between connecting capillaries appear to be due to the random nature of the network. The contribution of differences in the local hematocrit, the leukocyte obstruction, and the so-called apparent viscosity of blood to the pressure-flow relationships has not been established. Similarly, on the postcapillary side of the microcirculation, the physical dimensions of the system of collecting vessels are the primary mechanism affecting the continued loss in hydraulic pressure.

The intermittent nature of the red blood cell movement through the capillary network that has been so strikingly demonstrated by direct microscopy has led to the assumption that capillary pressure might likewise be continuously fluctuating. Landis (3) encountered a wide range of pressure values in what appeared to be identical vessels in the mesentery of the frog. In the present study, \(P_c\) was not measured routinely by direct intubation of the vessel but was measured at the junctional point of a side branch. In such instances, in what might be termed the midcapillary bed, time-averaged \(P_c\) fluctuated at most by 2-4 mm Hg, despite the erratic appearance of the red blood cell flow. Substantial changes in \(P_c\) developed in two ways. In one, the feeding precapillaries and the arterioles dilated spontaneously, causing a temporary increase in \(P_c\) by as much as 30-35%; however, \(P_c\) then returned to control levels after 5-7 minutes. Such excursions could occur several times during a 45-60-minute period. Conversely, the feeding terminal arterioles and precapillaries could constrict and stop capillary flow completely. The \(P_c\) in these instances fell to venular levels. Such interruptions in capillary flow persisted at most for 60-90 seconds and occurred irregularly with no discernible pattern. Throughout most of the observation period, \(P_c\) remained quite constant. In view of the passive contribution of the true capillaries and the postcapillaries to local regulation, active adjustments of pressure appear to reside primarily in the immediate precapillary vessels.

It is not within the scope of the present paper to discuss the question of shunting within the microcirculation. Suffice it to point out that, in the many networks which were studied, there were consistently one or two pathways from arteriole to venule which offered less overall resistance to flow so that the pressure drop along these capillaries was smaller than that in other parts of the network. In experiments on acute hemorrhage in cats leading to sustained hypotension (central arterial pressure 40-45 mm Hg), these same vessels in the mesenteric bed maintained a slowed but continuous flow of blood, even though other capillaries had a sluggish flow or were stagnant (unpublished observation).

An attempt was made to provide a definitive evaluation of average \(P_c\) calculations. Averages derived by four different procedures through direct microscopy gave reasonably similar values for the mesentery. The statistical mean value for circumscribed modules in the mesentery probably represents the most accurate estimate which uses simple numerical analysis. Averages based on the pressure drop between the arteriole and the venule tend to have wider extreme values. Such average pressures are most instructive when they represent a population of vessels whose physical characteristics (length, size, number, branching sequence) serve as a frame of reference for a more rigorous analysis. The presence of shunting vessels will, however, bias such averages. Estimates of \(P_c\) obtained by venous wedge techniques provide only an approximation of postcapillary pressures.

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References


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BENJAMIN W. ZWEIFACH

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