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The Effect of Changes in Arterial pCO_2 on Isogravimetric Capillary Pressure and Vascular Resistances* (33991)

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Total and prolonged arrest of the circulation can result in an increased vascular permeability (1–3). Among the factors that might change capillary permeability under the condition of absent or reduced blood flow, the first to be considered are those associated with continued metabolism of tissues such as reduced oxygen tension, local pH changes due to accumulation of metabolites such as lactic acid, and increased pCO_2.

The primary object of the present investigation was to determine if changes in carbon dioxide tension causes an alteration in capillary permeability in the canine forelimb. In studies on single mesenteric capillaries of frogs, Landis (4) found that changes in pCO_2 within the physiological range do not affect capillary permeability. Since no studies have been carried out in a mammalian species, it was considered to be of value to devise experiments using a method which permitted measurement of changes not only in a single capillary but in a complete organ and to also determine the effects of changes in pCO_2 on pre- and postcapillary vascular resistances.

Methods. Experiments were performed on mongrel dogs of either sex anesthetized with sodium pentobarbital, 30 mg/kg of body weight. Sixteen experiments were carried out in which one forelimb of a small dog (5–7 kg of body wt) was surgically removed above the elbow. Leg tissue was severed between double ligatures. Cutaneous bleeding was controlled by cautery and that from marrow cavity stopped with bone wax. All veins were dissected free for several centimeters above the joint. All animals were given heparin (5 mg/kg of body wt) at completion of surgery.

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The brachial artery and brachial and cephalic veins were cannulated with plastic tubing and the venous outflow was combined using a Y-piece. The isolated forelimb was placed on the platform of a continuously recording strain gauge weighing device. To perfuse the isolated leg, the left lung and the heart of another dog was removed and the pulmonary artery and the left atrium were cannulated. The heart was tied off around the ventricles. The trachea was intubated and attached to a Harvard respirator. The gas mixtures were delivered through a 5-liter reservoir bag connected to the inspiratory port of the respirator. The blood of a third dog was used to prime the perfusion system. From an elevated reservoir, blood flowed by gravity into the lung via the pulmonary artery. Blood flow was regulated by a screwclamp placed on the lung inflow tubing and adjusted to a pressure of 5–15 mm Hg measured in the left atrium. Blood from the left atrium was perfused through the forelimb by a Sigmamotor pump with a constant flow rate, excess flow returned to the reservoir by a Y-piece. The flow to the limb was adjusted by means of a screwclamp. Venous return was collected in a reservoir in a constant temperature water bath and returned to the elevated reservoir by means of a Sigmamotor pump. In addition, blood was circulated from the upper reservoir through a plastic tubing to the lower reservoir to assist in maintaining constant blood temperature. The total priming volume of the system was about 1000 ml. Pressures in the cephalic and brachial veins were regulated by an adjustable clamp, and the venous outflow was measured with a stopwatch and graduated cylinder. Arterial and venous pressures in the forelimb, and the pressure in the left atrium were monitored by means of needle-tipped catheters attached to Statham pressure transducers. The pressures and changes in leg weight were continuously registered on a Sanborn six-channel recorder. The temperature of the blood was monitored before entering the forelimb by means of a mercury thermometer.

The capillary permeability determination was a modification of the Pappenheimer and Soto-Rivera method (5). In this technique the pressures and flow are adjusted to a balance so that no net weight changes occur in the forelimb, that is an isogravimetric state. Initial flow rates were adjusted to the highest values at which the leg remained in the isogravimetric state and the venous pressure was maintained between 0.5 and 2 mm Hg. Following a period of equilibration, blood flow was decreased to about 50% of the initial flow and venous pressure adjusted to maintain the isogravimetric state. Then blood flow was decreased to a very low rate (1–2 ml/min) and venous pressure readjusted to maintain isogravimetricity. As a final step, the arterial inflow and venous outflow were simultaneously occluded as proposed by Johnson *et al.* (6). Since the pressure should equalize throughout all portions of the vasculature at zero flow, the isogravimetric capillary pressure (Pc_i) could be obtained by measuring the isogravimetric venous pressure (Pv_i). Initial flow rates were reestablished following each zero flow period. Each set of values was established after a period of approximately 5 min duration during which the foreleg weight remained constant. After 4 hr of perfusion, each experiment was terminated to avoid a possible decrease of the Pc_i resulting from deterioration of the preparation.

Different pCO_2 values were obtained by separately ventilating the isolated lung with three different gas mixtures of 5% CO_2, 25% O_2, 70% N_2; 10% CO_2, 25% O_2, 65% N_2; and pure oxygen without CO_2. Sequences of the administered gas mixtures were randomized. Ventilation was maintained for 20 min after each change in the gas mixture. A blood sample was then taken from the tubing leading to the brachial artery and analyzed for pCO_2, pH, and pO_2 on an Instrumentation Laboratory blood gas analyzer (model 1 113-S1) at 37°. Since the blood temperature throughout the experiment was maintained constant at 37°, temperature corrections were not necessary. After each change of the gas mixture, Pc_i measurements were repeated using the original gas mixture. Arterial and venous vascular resistances were calculated in

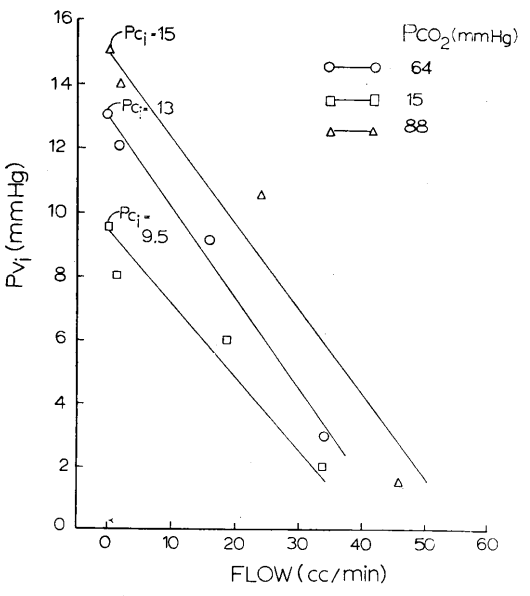

FIG. 1. Effect of changes in arterial pCO_2 on relationship between Pv_i and blood flow, and on Pc_i in a single foreleg experiment. At zero flow, Pv_i equals Pc_i.

isogravimetric periods under conditions of highest flow rate.

Results. Effect of changes in inspired CO_2 on arterial pCO_2 and pH. Figure 1 illustrates the determination of the Pc_i with different CO_2 tensions. Pv_i shown at zero flow, at which time the pressure is the same throughout the vasculature equals the Pc_i. Changes of the Pc_i associated with the alteration in pCO_2 shown in Fig. 1 is representative of the results of this investigation.

In Table I, effects of different CO_2 concentration in the ventilation gas mixture on pCO_2, pH, and Pc_i in each single experiment are listed. In the first three experiments, room air was used to obtain a low pCO_2, while in all succeeding studies pure oxygen was used to avoid hypoxemia. There was no evidence that the pO_2 in the range used in the present paper (51–530 mm Hg) had any influence on the behavior of the Pc_i.

If the Pc_i of single experiments are compared to each other after stepwise increases of pCO_2, there is a consistent decrease in Pc_i at low pCO_2 values compared to that seen at high pCO_2 levels. An unchanged or increased Pc_i with one exception is found, *i.e.*, Expt. 6 in which the increase of the pCO_2 from 53 to 73 mm Hg resulted in a fall of the Pc_i from 14 to 13.5 mm Hg.

The blood pH values at the 5% CO_2 ventilating gas mixture reveal a mean value of 7.2 indicative of metabolic acidosis. Since mean blood pCO_2 was 48 mm Hg, this degree of acidosis may be explained by an accumulation of fixed acids originating from the continued blood and tissue metabolism in the absence of liver and kidneys.

Table II summarizes the mean alterations of pCO_2 and the simultaneous mean changes in Pc_i. The statistical analysis of the changes in Pc_i demonstrate significant differences ($p = 0.05$) (Sign test).

Effect of pCO_2 on vascular resistances. Knowing the Pc_i, the total vascular resistance during each isogravimetric period may be fractionated into its arterial and venous components according to the formulas:

$$Rv = (Pc_i - Pv_i)/F,$$
$$Ra = (Pa_i - Pc_i)/F.$$

Rv = venous resistance; Ra = arterial resistance; Pv_i = isogravimetric venous pressure; F = flow; Pa_i = isogravimetric arterial pressure.

In Table III, changes in pre- and postcapillary resistances after the different CO_2 levels are shown. On the arterial side, the reaction of the resistance is consistent: in all experiments, the precapillary vascular bed dilated with increased CO_2. The behavior of the venous vessels does not seem to follow a uniform pattern. Changes in pCO are associated with increased as well as in decreased venous resistance.

Discussion. The present study suggests a significant relationship between blood pCO_2 and capillary permeability. The question of whether the primary responsible agent is carbon dioxide or hydrogen ion is not resolved from these studies. Landis (4) bathed the exposed mesentery of frogs in Ringer's solution equilibrated with various amounts of CO_2, used sodium bicarbonate to keep the pH between 5.1 and 5.4, and stated the following: "Changes in carbon dioxide tension which lie within the physiological range do

TABLE I. Effect of Changes in Inspired CO_2 on pCO_2, pH, and Pc_i in the Isolated Canine Forelimb.

	Inspired CO_2 (%)											
	0				5				10			
Expt. no.	pCO_2 (mm Hg)	pH	pO_2 (mm Hg)	Pc_i (mm Hg)	pCO_2 (mm Hg)	pH	pO_2 (mm Hg)	Pc_i (mm Hg)	pCO_2 (mm Hg)	pH	pO_2 (mm Hg)	Pc_i (mm Hg)
1	10	7.53	63	10.0	46	7.20	117	12.5	—	—	—	—
2	10	7.83	51	8.5	—	—	—	—	71	7.08	119	10.5
3	10	7.55	65	10.5	37	7.28	60	11.5	—	—	—	—
4	14	7.57	180	12.0	41	7.21	110	17.0	—	—	—	—
5	15	7.42	340	12.5	52	7.27	200	14.0	71	7.14	180	18.5
6	15	7.32	350	12.5	53	7.19	165	14.0	73	7.11	160	13.5
7	15	7.44	515	9.5	64	7.08	157	13.0	88	7.04	165	15.0
8	12	7.57	450	6.5	41	7.26	150	10.0	—	—	—	—
9	9	7.58	465	10.5	41	7.04	165	10.5	68	7.04	155	12.0
10	10	7.45	480	7.0	47	7.06	176	10.0	86	6.95	170	10.0
11	17	7.34	480	12.0	57	7.12	170	12.0	71	7.00	152	14.0
12	—	—	—	—	46	7.14	165	16.0	70	7.01	135	20.0
13	—	—	—	—	50	7.14	205	15.5	90	7.00	175	16.5
14	10	7.48	530	10.0	55	7.14	210	15.0	—	—	—	—
15	—	—	—	—	51	7.21	180	11.0	100	6.94	170	14.0
16	—	—	—	—	43	7.14	165	13.0	80	6.90	200	17.5
Mean	12	7.51	330	10.1	48	7.16	159	13.0	78	7.02	162	14.7
SD	6.7	0.13	189.3	2.0	7.2	0.06	39.1	2.2	10.6	0.06	21.9	3.1

TABLE II. Comparison of Mean Changes in Blood pCO_2 on Pc_i in the Isolated Canine Forelimb.

Mean change in blood pCO_2 (mm Hg) ± SE	12.0 (±1.9) to 48 (±1.8)	48 (±1.8) to 78 (±3.2)	12.0 (±1.9) to 78 (±3.2)
Mean change in Pc_i (mm Hg) ± SE	10.1 (±0.6) to 13.0 (±0.5)	13.0 (±0.5) to 14.7 (±0.9)	10.1 (±0.6) to 14.7 (±0.9)
No. of expts.	11	10	7
Statistics on change in Pc_i[a]	$p = 0.004$	$p = 0.04$	$p = 0.016$

[a] Sign test.

TABLE III. Effect of Changes in Inspired CO_2 on Pre- and Postcapillary Resistances in the Isolated Canine Forelimb.

Expt. no.	CO_2 (%):	Precapillary resistance (mm Hg/ml/min) 0	5	10	Postcapillary resistance (mm Hg/ml/min) 0	5	10
1		8.00	3.40	—	0.50	0.47	—
2		6.94	—	4.51	0.27	—	0.37
3		10.02	7.44	—	0.18	0.78	—
4		13.36	4.10	—	0.29	0.35	—
5		2.30	2.00	1.80	0.40	0.33	0.30
6		4.80	4.40	1.90	0.64	0.64	0.34
7		1.60	1.10	0.98	0.11	0.20	0.27
8		2.12	1.29	—	0.06	0.19	—
9		5.40	4.40	1.70	0.18	0.56	0.32
10		0.90	0.70	0.69	0.06	0.09	0.09
11		4.73	3.32	2.31	0.25	0.37	0.35
12		—	4.55	1.86	—	0.44	0.21
13		—	1.33	1.21	—	0.22	0.16
14		2.40	1.70	—	0.18	0.29	—
15		—	8.00	6.17	—	0.50	0.36
16		—	3.68	2.04	—	0.59	1.18

not influence the movement of fluids by any direct effect upon capillary permeability but only in an indirect manner through the rise in capillary pressure which accompanies hyperemia." The fluid shift following hyperemia, which is described by Landis, corresponds to the first factor of Starling's hypothesis of a transcapillary fluid exchange (7). According to this hypothesis, three factors are responsible for the transcapillary fluid exchange: (i) the difference in hydrostatic pressure across the capillary membrane; (ii) protein osmotic pressure acting within the vascular bed and in the extravascular compartment; and (iii) the properties of the capillary membrane which influence transfer from intravascular to extravascular region. The vasodilating effect of CO_2 which Landis could observe in his study is confirmed by the findings of Daugherty *et al.* (8) and Fleisch *et al.* (9) in the dog forelimb and the cat hindlimb, respectively, and by the results of the present experiments.

The fractionation of the total vascular resistance into the precapillary and postcapillary portions showed a consistent and marked fall of the arterial resistance, but the postcapillary resistance was not altered consistently. The action of CO_2 on the postcapillary vessels is perhaps of a more indirect nature. If a rise in pCO_2 with vasodilation and drop in arterial pressure caused a diversion of blood flow from previously existing A–V shunts to the capillaries, an increasing venous resistance could result. On the other hand, an arterial–venous reflex, tending to keep the capillary pressure constant, could cause the venous resistance to decrease after the rise in capillary inflow due to increased pCO_2. However, Johnson *et al.* (6) did not find autoregulation in the forelimb of the dog.

Since the arterial resistance decreased and the venous pressure did not change consistently, it can be concluded that the capillary hydrostatic pressure in the present experiments rose when pCO_2 increased. But capillary filtration represented by a slow, progressive weight change could not be observed. There was an initial weight increase which, however, can be explained by an increase in blood volume after vasodilation.

The lack of capillary filtration was, however, associated with an increased Pc_i, which can be defined as the sum of all pressures opposing filtration. The most important force

opposing filtration is the effective colloid osmotic pressure within the capillaries, which is the difference between osmotic pressure of intra and extravascular fluids. The protein osmotic pressure depends on the protein concentration, on the ratio of globulin to albumin and on the pH. Since in the present studies changes in pCO_2 were accompanied by pH alterations, there could have been a direct effect on the colloid osmotic pressure. The pH effects, however, are very small so that Pappenheimer (5) could not detect a change in osmotic pressure when pH was changed from 7.2 to 7.7.

The explanation of the changes in Pc_i which seems to have the highest degree of likelihood involves the third factor of Starling's hypothesis: the properties of the capillary membrane. The physical properties of the capillary membrane influence the balance of intra- and extravascular protein concentration and therefore the effective colloid osmotic pressure in the capillary. The permeability of the capillaries was clearly demonstrated by several authors (10–12). If a given pCO_2 modifies the capillary membrane, altering the balance between intra- and extracapillary colloid osmotic pressure, the Pc_i could be changed. According to this thesis, the capillary membrane is actively involved in a mechanism which is responsible for a steady state in the transcapillary fluid exchange. A rise in capillary hydrostatic pressure following an increased pCO_2 would be balanced to a certain degree by an increased effective capillary colloid osmotic pressure due to changes in the physical properties of the capillary membrane.

The present study shows the effect of pCO_2 values in blood, which lie within a range compatible with life, on the mean capillary pressure and on pre- and postcapillary vascular resistances of the isolated dog forelimb. However, further work is indicated in order to apply these results to the intact animal preparation, especially in regard to the probable influence of generalized respiratory acidosis and the possibility of evoked activities of the sympathicoadrenal system on the vascular resistances and capillary blood pressure.

Summary. The study was designed to determine the effect of a changing arterial pCO_2 on capillary membrane permeability and pre- and postcapillary resistances. Experiments were carried out on isolated forelimbs of dogs perfused with blood equilibrated with different CO_2 pressures in an isolated lung. Gas mixtures containing 0, 5, and 10% CO_2 resulted in arterial mean pCO_2 values of 12, 48 and 78 mm Hg. Isogravimetric capillary pressure (Pc_i) was measured using a modified Pappenheimer and Soto-Rivera technique. Results show the mean Pc_i rising from 13 to 14.7 mm Hg ($p = 0.04$) after pCO_2 was increased from 48 to 78 mm Hg. When the pCO_2 was lowered to 12 mm Hg, Pc_i fell to 10.1 mm Hg ($p = 0.016$). Precapillary resistance fell with an increased pCO_2 and rose with a decreased pCO_2. Effects of pCO_2 alters the permeability of the capillaries due to a direct action on the capillary membrane. The observed changes in capillary permeability are interpreted as a protective mechanism opposed to effects of alterations of capillary hydrostatic pressure resulting from changes in blood pCO_2.

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