A STUDY OF THE MECHANISM BY WHICH EXERCISE INCREASES THE PULMONARY DIFFUSING CAPACITY FOR CARBON MONOXIDE* †

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The pulmonary diffusing capacity (D_L) increases with exercise. This was first shown by Krogh in 1915 (1) and has been confirmed by investigators who have studied the matter since that time, using both carbon monoxide and oxygen (2–5). The mechanism by which exercise increases the diffusing capacity is not altogether clear, however, and the apparent amount of increase with exercise is dependent on the technique used for determining the diffusing capacity, and on the severity of the exercise.

Two methods widely used for estimating the pulmonary diffusing capacity for carbon monoxide are the steady state method of Filley, MacIntosh and Wright (3) and the Krogh breath holding technique as modified by Ogilvie, Forster, Blakemore and Morton (5). In the steady state technique, the subject breathes continuously throughout the test period, which makes minute ventilation a variable factor. In the breath holding method, however, the breath is held and minute ventilation cannot be a variable factor during the actual test period.

The present study was undertaken to investigate the cause of the increased pulmonary diffusing capacity for carbon monoxide which occurs with exercise, using both the steady state and the breath holding techniques. Both methods were used in this study because both have been widely employed for the measurement of diffusing capacity and both show an increase in diffusing capacity with exercise, but the conditions of measurement are different enough to indicate that results with one of these methods might serve as a useful check on the reliability of conclusions reached by the use of the other method. The effects of altering pulmonary blood flow, ventilation, pulmonary vascular pressures, blood pH and blood gas contents, either alone or in combinations, have been studied in an attempt to simulate some of the physiological effects of exercise which might conceivably alter the diffusing capacity. The results indicate that the factors apparently responsible for the increase in pulmonary diffusing capacity with exercise may differ with the technique used.

METHODS

Subjects. The subjects studied were normal, male, graduate and medical students with an age range of 21 to 26 years and surface area range of 1.70 to 2.14 M.². All were able to carry out the experimental procedures without difficulty.

Diffusing capacity of the lung for carbon monoxide (D_L).
a) Steady state technique. Steady state determinations of the pulmonary diffusing capacity were made by the method of Filley and co-workers (3). An indwelling brachial artery needle, and, in those subjects where cardiac output was measured, a superior vena cava catheter, were inserted and the subject was allowed to lie quietly for 30 minutes before the studies were begun. He then inspired 0.1 per cent carbon monoxide in air through a Krogh valve for six minutes and the sample for measurement of D_L was collected during the last two minutes. For determination of D_L during exercise, the subject inspired air for the first two minutes and the 0.1 per cent carbon monoxide mixture for the last four minutes of the six minute period, with sample collection during the last two minutes. Expired gas was collected in a Douglas bag during the fifth and sixth minutes and the number of expirations for this two minute period was recorded. Injection of the T-1824 dye and collection of arterial blood samples at two second intervals for cardiac output determination were carried out as close to the initial part of the two minute collection period as was possible. This usually required 30 to 40 seconds. An arterial blood sample for the determination of pCO_2 was then withdrawn at a slow, regular rate into a heparinized syringe over the two minute period while expired gas was being collected.

Only one exercise determination and a total of no more than three D_L determinations were made in a single subject on any one day. The subject rested at least 20 minutes between each determination of D_L. Control studies in two subjects at rest showed no marked tendency for D_L to decrease progressively as the result of repeated measurements under these conditions. The successive values for D_L obtained with intervening recovery periods of 20 minutes were: for Subject D. G., 27.4, 26.9 and 25.0 ml. CO per minute per mm. Hg; and for Subject J. M., 33.1, 33.8, 33.0 and 32.6 ml. CO per minute per mm. Hg. This result indicated that the change in D_L caused by repeated measurements under these conditions would be small relative to the change produced by exercise. Because the emphasis in this study was on factors which cause a change in the diffusing capacity, rather than on the absolute value of the diffusing capacity, no attempt was made to correct for "back pressure" of carbon monoxide in the pulmonary capillary blood. The order in which D_L under different conditions was measured was randomized from subject to subject in order to minimize systematic error resulting from repeated measurement.

The volume of expired gas was measured in a Tissot gasometer and corrected to the volume at standard temperature and pressure, dry (S. T. P. D.). The expired gas was analyzed, immediately after collection, for carbon monoxide in an infrared analyzer[1] which gave results reproducible to 0.001 per cent carbon monoxide over the concentration range in use (0.000 to 0.100 per cent carbon monoxide). Gas samples for carbon dioxide and oxygen analysis were removed from the Douglas bag immediately after collection and stored over mercury in glass tonometers for analysis within four hours in the micro-Scholander apparatus. Analyses were done in duplicate and required to check within 0.1 per cent. The pH of whole blood was measured with a Cambridge Model R pH meter equipped with an enclosed glass electrode. Measurements were made at room temperature, usually 25 to 26° C., and the result was corrected to 37° C. by Rosenthal's factor (6). The per cent oxygen saturation of arterial blood was assumed to be 97 per cent for calculation of pCO_2 by nomogram. The arterial blood oxygen capacity was estimated from the total hemoglobin, which was determined by the spectrophotometric method of Drabkin and Austin (7). The carbon dioxide content of whole blood was determined by the method of Van Slyke and Neill (8), and duplicate analyses were required to check within 0.1 volume per cent. The plasma carbon dioxide content was estimated from the blood carbon dioxide content and from the pH, hemoglobin concentration and per cent oxygen saturation by the line charts of Van Slyke and Sendroy (9). The plasma CO_2 tension was calculated by the Henderson-Hasselbalch equation, using a pK' of 6.11 (10). In this laboratory, pCO_2 values by this method are reproducible to within ± 1.0 mm. Hg.

Calculations of D_L were made as outlined by Filley and co-workers (3):

$$\dot{V}_{CO} = \dot{V}_E \left(F_{ICO} \frac{F_{EN_2}}{F_{IN_2}} - F_{ECO} \right)$$

and

$$P_{ACO} = (B_p - 47) \frac{F_{ECO} - r F_{ICO}}{1 - r}$$

where

$$r = \frac{V_d' \text{ (physiological dead space volume)}}{V_t \text{ (tidal volume)}}$$

$$= \frac{P_{ACO_2} - P_{ECO_2}}{P_{ACO_2}}$$

$$D_L = \frac{\dot{V}_{CO}}{P_{ACO}}.$$

$\dot{V}_{CO}$ is the minute uptake of CO, $\dot{V}_E$ is the total gas volume expired per minute, F_I and F_E are the inspired and expired concentrations of the gases indicated by subscripts, P_{ACO} is the alveolar tension of CO, and P_{ACO_2} and P_{ECO_2} are arterial and expired tensions of CO_2, respectively.

When the inspired mixture contained 5.0 per cent CO_2,

$$r = \frac{P_{ACO_2} - P_{ECO_2}}{P_{ACO_2} - P_{ICO_2}}.$$

Since D_L is expressed as the quantity of CO diffused per minute per mm. Hg CO pressure gradient between alveolar gas and capillary blood, the correct equation for D_L is $\frac{\dot{V}_{CO}}{P_{ACO} - P_{CCO}}$, where P_{CCO} represents the mean carbon monoxide tension of pulmonary capillary blood. Filley and co-workers (3) regarded P_{CCO} as negligible and considered the pressure gradient substantially equal to the alveolar pressure of CO, thus omitting P_{CCO} from the equation for expediency. In the present study, P_{CCO} has been omitted from the equation and no correction for back pressure of CO was made in the calculations. This is not entirely correct for measurement of the absolute value of D_L, since it has been shown that P_{CCO} does effect steady state determinations (11, 12), and repeated steady state measurements might be expected to show progressive decline in apparent D_L because of the progressive accumulation of COHb. As pointed out previously, however, repeated measurements of D_L at intervals of 20 minutes in control subjects in the resting state indicated that the D_L was not being significantly altered, for the purposes of this study, by the changes in pulmonary capillary tension of CO brought about by preceding measurements.

Hyperventilation is one of the obvious consequences of exercise, and the effect on steady state D_L of hyperventilation without other exercise was therefore examined. The procedure for measurement of D_L by the steady state technique during hyperventilation requires description in some detail because of uncertainties which hyperventilation introduces into the method. The principal uncertainties are as follows:

1. The steady state method embodies the assumption that the physiological dead space and intrapulmonary distribution of carbon monoxide and carbon dioxide are similar, but it is uncertain how this relationship is affected by hyperventilation. However, the carbon monoxide dead space increases by about the same amount as that for carbon dioxide during the hyperpnea of exercise in normal persons (4), and it appears to be a reasonable assumption that this relationship also holds during voluntary hyperventilation.

2. Hyperventilation also imposes significant technical difficulties: a) As the difference between Pa_{CO_2} and PE_{CO_2} is progressively decreased by hyperventilation on air or, especially, on 5.0 per cent CO_2, an error in the determination of either Pa_{CO_2} or PE_{CO_2} will cause a progressively greater error in the determination of "r" and, consequently, in D_L. b) Failure to achieve a steady state may result in sampling which is unrepresentative of the entire test period with consequent possibility of large errors. In particular, overestimating mean Pa_{CO_2} by about 4 mm. Hg during hyperventilation on air could cause an apparent increase in D_L equal to that reported in Table V as resulting from hyperventilation. Underestimating Pa_{CO_2} causes D_L to be underestimated, but the absolute error is less than that caused by overestimation of Pa_{CO_2}. c) Failure to reach a steady state can cause some error even though ventilation rate and cardiac output are constant and arterial blood is sampled at a constant rate throughout the period of collecting expired air. This is because the CO_2 dissociation curve is not a straight line, so that the pooled arterial sample, although it represents accurately the mean CO_2 content of blood when this is changing during the collection period, may not represent exactly the mean CO_2 tension. Sample calculations indicate, however, that the error on this account is negligible for present purposes.

Because of these uncertainties, care was taken to keep hyperventilation as regular as possible. The frequency of respiration was timed with a metronome at 22 breaths per minute and the subjects were asked to keep tidal volume as constant as possible. Hyperventilation was maintained on the carbon monoxide mixture for six minutes, during the last two minutes of which expired gas was collected and arterial blood was continuously drawn at a steady rate for the determination of D_L. This procedure was carried out for hyperventilation on 0.1 per cent CO in air as well as 0.1 per cent CO in a mixture with 5.0 per cent CO_2. During these few minutes a condition of complete equilibrium was certainly not attained. For the six subjects of Table V, the control respiratory exchange ratio was 0.96 (S.D., 0.13) and that during hyperventilation on air was 1.33 (S.D., 0.20). For the three subjects who also hyperventilated on 5.0 per cent CO_2, the values at this time were 0.36, 0.51 and 0.67. The question of importance for the present use of the steady state method, however, is the extent to which conditions were stable during the test period. To examine this question, end-expiratory alveolar air samples were collected from the first three subjects at the end of each test period and the PA_{CO_2} calculated from these samples was compared with the mean Pa_{CO_2} of the arterial blood sample which was drawn continuously through the two minutes of the test period. For the resting state, the values of arterial minus alveolar pCO_2 were −0.1, 0.9 and −0.6 mm. Hg; for hyperventilation on air, they were 1.9, −0.1 and 1.2 mm. Hg; and for hyperventilation on CO_2, they were −0.2, −0.2 and 0.2 mm. Hg. These results indicate that conditions during the test period were adequately steady for use of the method. As an additional check, the oxygen contents of inspired, expired and alveolar air were used for an independent calculation of "r" and D_L. This precaution is especially desirable in experiments dealing with hyper-

ventilation on CO_2, since analytical errors in CO_2 measurements here can cause large errors in estimation of D_L. As an auxiliary test gas, oxygen also has the advantages of being somewhat less sensitive than CO_2 to variations in the steady state, and of having physical properties close enough to carbon monoxide to make it probable that intrapulmonary distribution of the two gases during hyperventilation is similar. Results with the two test gases were quite similar, as shown in Table V, which presents D_L values obtained during the same experiments using arterial pCO_2, alveolar pCO_2 and alveolar pO_2. These all show substantial agreement.

These experiments indicate that the steady state method can be used satisfactorily during hyperventilation under the conditions of this investigation.

b) Breath holding technique. Measurements of the breath holding D_L were made by the modification of the Krogh technique described by Forster and co-workers (5, 13). Using this technique, the subject made a maximal inspiration from the level of residual volume, held his breath for approximately 10 seconds with the glottis open, avoiding a valsalva maneuver, and then rapidly expired, delivering the entire expirate within two seconds. All of the expired air except the first liter, usually amounting to between 2.5 and 3 L., was collected to be analyzed as alveolar gas. The inspired gas mixture contained 0.3 per cent carbon monoxide, 10 per cent helium and approximately 20 per cent oxygen in nitrogen and was inspired from a bag in a box connected with a Collins spirometer so that the inspired volume was recorded on the spirometer. The exact duration of breath holding, timed from the beginning of inspiration to the start of collection of the alveolar sample, was recorded on the spirometer kymograph drum, which was turning at a constant rate. The alveolar sample was collected through a stopcock in the circuit near the subject's mouth. After the subject expired approximately 1 L. (as observed on the spirometer tracing), which was considered more than adequate to wash out the dead space, the stopcock was turned so that the remainder of the expirate was diverted into a collapsible plastic bag, previously evacuated to low pressure by a pump. Residual volume was determined in each subject by the open circuit helium method of Hickam, Blair and Frayser (14) and, since inspiration was made from residual volume, alveolar volume was taken as the sum of residual volume and inspired volume.

The alveolar gas samples were analyzed for helium, carbon monoxide and oxygen. The helium concentration was determined by katharometer[2] with an accuracy of about one part in 100 or 0.05 per cent helium (14). Carbon monoxide concentration was measured by the infrared analyzer with an accuracy of about one part in 100 or 0.001 per cent carbon monoxide. Analyses for helium and carbon monoxide were made after removal of CO_2 by passage through "ascarite." The helium and carbon monoxide concentrations obtained in this manner were then corrected for the volume change resulting from removal of carbon dioxide prior to analysis. In order to make this correction, determination of carbon dioxide con-

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[2] Hastings Instrument Co.

centration in the alveolar sample was required. This was done indirectly as follows. The oxygen concentration in the dried gas sample was determined by a Pauling analyzer,[3] both before and after removal of carbon dioxide by passage through "ascarite." The carbon dioxide concentration could then be estimated by the change in oxygen content of the gas sample resulting from carbon dioxide absorption. Change in oxygen concentration can be measured, under these conditions, with an accuracy of 0.1 per cent. Estimate of carbon dioxide by this means is relatively crude, but is accurate to less than 0.5 per cent. This degree of inexactitude introduces an error of less than one part in 200 in the estimate of carbon monoxide and helium concentrations. In the helium analysis, allowance was made for the effect on the katharometer of changes in the oxygen-nitrogen ratio in the expired gas from the value

at which the katharometer was calibrated (0.26). A decrease in the ratio caused an apparent decrease in helium concentration as measured by the katharometer, amounting to 0.17 per cent helium for a change in ratio from 0.3 to 0.2. The appropriate correction was made in each case.

For the studies in which the cardiac output was measured, the indwelling brachial artery needle and superior vena cava catheter were put in place at least 30 minutes before the tests were begun. At least 10 minutes were allowed between each determination of D_L by the breath holding method.

The carbon monoxide concentration present in the alveolar sample before any had been absorbed by the lungs was calculated from the dilution of helium by the equation of Forster and co-workers (5, 13):

$$\text{Initial CO concentration in the expired alveolar sample} = \frac{\text{He concentration in the expired alveolar sample}}{\text{Inspired He concentration}} \times \text{Inspired CO concentration.}$$

D_L was then calculated from Krogh's equation (1):

$$D_L = \frac{\text{Alveolar volume (S.T.P.D.)} \times 60}{\text{Time (seconds)} \times (Bp - 47)} \times \text{Natural log} \frac{\text{Initial CO concentration in the expired alveolar sample}}{\text{Final CO concentration in the expired alveolar sample}}$$

with D_L expressed as ml. CO (S.T.P.D.) diffused per minute per mm. Hg CO tension.

Measurement of cardiac output and central venous pressure. Cardiac output was determined by the dye dilution technique (15), with central venous injection of the dye as described by Doyle, Wilson, Lepine and Warren (16), using T-1824. A No. 6 calibrated catheter was inserted through an antecubital vein and the end placed in the superior vena cava by fluoroscopic observation so that T-1824, in 0.5 per cent solution, could be injected from a calibrated syringe into the central circulation. After the dye was injected, arterial blood samples were collected in 3 ml. siliconized tubes at two second intervals. The undiluted dyed serum from each two second sample was read on a Beckman DU spectrophotometer against a serum blank. The optical densities were plotted against time on semilogarithmic graph paper and all the points from the first appearance of the dye in the serum until recirculation began were used in construction of a curve. With exercise and with the cardiac output increased by drugs, from five to eight points were available for constructing the curve. Under all other conditions, from seven to 11 points were used. Calculations of the cardiac outputs were then made from the time-concentration curves (16).

Since the purpose of cardiac output measurements was to study the relationship of cardiac output to D_L, it would have been desirable to make the measurements of cardiac output at precisely the same time D_L was being determined. This, however, would have required a second arterial puncture for the steady state procedure since an arterial blood sample for pCO_2 determination was being withdrawn continuously throughout the two minute D_L determination period. Dye injections and arterial sample collections for cardiac output determinations, therefore, were carried out immediately before the arterial blood for pCO_2 was taken.

When D_L was determined by the breath holding technique, it was again technically not feasible to collect the arterial blood samples for cardiac output measurement at the precise time D_L was being measured. For this reason, the procedures for measuring cardiac output were carried out in the 30 to 40 seconds immediately before D_L determination. In order to maintain conditions comparable to those during the measurement of D_L, the breath was also held with glottis open during the injection of dye and the collection of arterial blood samples. The same technique was used each time a cardiac output was measured together with a breath holding D_L under various experimental conditions.

The central venous pressure was measured by way of the superior vena cava catheter, using a Statham strain gage (0 to 25 mm. Hg), and was recorded by a direct writing recorder. The point of zero reference was arbitrarily taken at a position midway between the table surface and the fourth costochondral junction. The matter of primary interest was change in pressure rather than absolute value of pressure.

Exercise. Exercise was carried out while supine by having the subjects alternately flex and extend the legs rapidly, sliding the heels along the table top. The subjects exercised for six minutes for both steady state and breath holding studies.

Procedures for increasing cardiac output without exercise. Cardiac output was increased by drugs in six subjects studied by the steady state method and in 12 subjects studied by the breath holding technique. In three subjects of the steady state group, the cardiac output was increased by epinephrine, 0.17 to 0.19 μg. per Kg. per minute, given by intravenous infusion over a 10 minute period before the D_L and cardiac output were determined and maintained during these determinations. In the other

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[3] Model E-2, Arnold O. Beckman Inc.

TABLE I

Steady state D_L, cardiac output and ventilation rate, in 19 normal supine subjects at rest

Subject	Age	Surface area	Number of determinations	Ventilation rate*	Cardiac output	D_L
		$M.^2$		$L./min.$	$L./min.$	$ml./min./mm. Hg$
D. P.	22	1.86	1	8.1	7.8	24.4
R. D.	25	1.96	2	7.4	6.1	32.4
				6.5	6.8	23.5
F. D.	24	2.10	2	6.3	7.5	24.6
				6.7	9.3	23.5
R. R.	24	2.14	1	8.8	6.7	21.9
S. S.	22	1.70	2	7.8	6.4	25.1
				5.3	4.2	19.6
C. R.	20	1.82	2	4.7	6.7	23.9
				6.1	9.4	21.7
H. S.	22	1.72	2	5.8	10.0	22.6
				7.4	6.4	22.0
S. W.	23	1.97	1	4.7	6.1	27.2
D. G.	22	2.06	1	6.5	7.1	19.9
C. W.	24	1.87	1	5.0	5.4	24.5
B. S.	21	1.88	1	5.4	6.2	24.6
R. R.	26	1.75	1	6.8	6.7	19.9
S. R.	22	1.70	1	7.4		22.0
B. C.	23	2.00	1	7.6	6.1	24.3
B. M.	22	1.96	1	4.9	5.8	21.3
J. F.	23	1.86	1	5.8	6.0	19.4
J. S.	21	2.08	1	8.7	10.3	29.2
F. A.	21	1.76	1	7.6	7.7	22.9
J. B.	21	2.12	1	7.1	9.4	23.6
Mean	22.5	1.91		6.6±1.2†	7.0±1.4	23.5±3.0

* All ventilation rates are in L. per minute S.T.P.D.
† Values following the means are standard deviations.

three subjects of the steady state group, and in all the 12 subjects where breath holding D_L was determined while the cardiac output was increased by drugs, the cardiac output was increased by a combination of atropine sulfate and norepinephrine, which has also been found effective in increasing cardiac output by Bryant, Combs and Warren (17). Atropine sulfate (2.0 mg.) was given intravenously over a two minute period followed by an intravenous infusion of norepinephrine given at the rate of 8.0 μg. per minute over a six minute period before the determinations of D_L and cardiac output were made and maintained during these determinations.

In some experiments, the cardiac output was increased by producing reactive hyperemia in the legs with arterial tourniquets (18). The tourniquets were 6 inch sphygmomanometer cuffs which were placed around the upper thighs, inflated, and maintained at a pressure of 200 mm. Hg for 15 minutes. The pressure was relieved suddenly by an escape valve. This procedure, which acutely reduces peripheral resistance, will approximately double the resting cardiac output for about a minute.

Procedures intended to produce pulmonary vascular engorgement. The effect of procedures designed to increase the pressure in the pulmonary capillary bed on the steady state D_L was studied in several subjects. The procedures used were rapid expansion of the plasma volume by intravenous infusion of human serum albumin solution and reduction in size of the peripheral vascular bed by norepi-

⁴ Made by the David Clark Co., Inc., Worchester, Mass.

nephrine infusion or by compression of the body by an aviator's "G suit."⁴ Pulmonary vascular pressures were not measured. It has been found, however, that these procedures will increase central venous pressure in normal subjects by about the same amount that they increase pulmonary capillary and pulmonary artery pressures. Changes in central venous pressure (CVP) were therefore used as a rough index of change in pulmonary vascular pressures. The support for this concept is provided by the work of several different investigators. Doyle, Wilson, Estes and Warren found that pulmonary arterial and capillary pressures were increased by rapid infusion of saline solution (19) to about the same extent that such infusions increase central venous pressure (20). They concluded that the rise in pressure produced in this way is essentially the same throughout the central veins and pulmonary vessels, with maintenance of normal pressure gradient at a higher level. Fowler, Westcott, Scott and McGuire (21) found that norepinephrine infusion produced a rise in pulmonary capillary pressure, and, in the three subjects where it was measured, a similar increase in right atrial pressure. Wood, Lewis, Forster and Beckman (22, 23) found that inflation of a "full pressure half-suit" around the lower half of the body produced similar increases in central venous, right heart, pulmonary artery and pulmonary capillary pressures.

In the present study, four subjects had an infusion of between 400 and 500 ml. of 5.0 per cent serum albumin in physiological saline over a six minute period to produce

pulmonary vascular engorgement while the steady state D_L was being determined.

In three subjects, inflation of the G suit over the lower half of the body was used alone to produce pulmonary vascular engorgement. An attempt was made to keep the suit inflated at high pressure (two pounds per square inch) to cause marked increase in the CVP for the entire six minute period of the steady state D_L measurement. This was uncomfortable, however, and therefore the pressure during this time was alternately applied and released at 15 second intervals.

Pulmonary vascular engorgement was produced in two subjects by a combination of norepinephrine infusion with G suit inflation. When the suit was inflated to 1.5 pounds per square inch, it was found that the CVP was at first elevated but then decreased gradually even though G suit pressure was maintained. Bondurant, Hickam and Isley (24) observed this previously and used norepinephrine infusion to maintain the increase in CVP produced by G suit inflation. For this reason, norepinephrine infusion was added to G suit inflation in these two subjects, and the CVP remained elevated throughout the six minute period.

In six subjects, a combination of G suit inflation (1 to 1.5 pounds per square inch) and infusion of 300 to 400 ml. of 5.0 per cent serum albumin solution in saline over the six minute D_L determination period was used to cause pulmonary vascular engorgement. With this method, high central venous pressures were produced and maintained for the six minute period without causing significant discomfort.

RESULTS

A. Studies with D_L determined by the steady state method

The studies in normal resting subjects are summarized in Tables I and II. Nineteen supine subjects had a mean D_L of 23.5 (SD ±3.0) ml. per minute per mm. Hg. Six other subjects in the seated position had a mean D_L of 25.5 ± 5.3 ml. per minute per mm. Hg. In this group of subjects, the differences between seated and supine D_L values are not significant.

TABLE II

Steady state D_L, ventilation and cardiac output in six normal seated subjects at rest

Subject	Age	Surface area	Ventilation rate	Cardiac output	D_L
		$M.^2$	$L./min.$	$L./min.$	$ml./min./mm. Hg$
D. J.	21	1.84	6.8	5.3	22.0
F. D.	24	2.14	5.8	7.2	23.5
C. H.	21	2.14	7.8	4.5	26.7
R. C.	23	2.02	7.9	8.4	35.8
B. L.	23	2.14	8.3	7.6	23.1
J. C.	23	1.79	6.5	9.5	22.0
Mean	22.5	2.01	7.2±0.9	7.1±1.9	25.5±5.3

Effect of exercise and increased cardiac output on the steady state D_L

Cardiac output and D_L were measured simultaneously in six subjects, first at rest, then during exercise, and finally during a period when the cardiac output was increased by drugs while the subjects were otherwise at rest, as summarized in Table III (see Methods). With exercise, the mean cardiac output increased to 13.6 ± 3.9 L. per minute from the resting value of 7.7 ± 1.6 L. per minute, while the mean D_L increased to 39.8 ± 5.5 ml. per minute per mm. Hg from the resting mean of 25.4 ± 3.6 ml. per minute per mm. Hg. When the cardiac output was comparably increased by drugs without change in the mean ventilation rate, there was no increase in D_L over the resting level.

In four subjects, the intravenous infusion of 400 to 500 ml. of 5.0 per cent human serum albumin in normal saline increased the mean cardiac output to 10.7 ± 1.2 L. per minute from 6.7 ± 2.6 L. per minute at rest before the infusion, but the mean D_L remained substantially unchanged (Table IV).

In summary, exercise caused a striking increase in steady state D_L, but increasing the cardiac output to the same extent by drugs or albumin infusion did not change D_L.

Effect of increased ventilation rate on the steady state D_L

The effect of voluntary hyperventilation, carried out as previously described, on the steady state D_L was studied in six subjects who were otherwise at rest (Table V). The mean ventilation rate changed from 6.1 ± 0.9 to 16.0 ± 3.6 L. per minute, while the mean cardiac output remained essentially the same as at rest. The mean D_L increased from the resting level of 22.3 ± 2.2 ml. per minute per mm. Hg to 38.3 ± 7.1 ml. per minute per mm. Hg with hyperventilation.

The first three subjects shown in Table V hyperventilated while breathing 0.1 per cent carbon monoxide in a mixture with 5.0 per cent carbon dioxide as well as while breathing 0.1 per cent carbon monoxide in air. Hyperventilation while breathing the carbon dioxide mixture prevented the arterial pH and pCO_2 changes which occurred with hyperventilation on air, but the

TABLE III

Steady state D_L, ventilation rate, cardiac output and oxygen uptake, in six supine subjects at rest, during exercise, and with the cardiac output increased by drugs

Subject	Condition	M.V.*	O₂ uptake	C.O.†	D_L
		L./min.	*ml./min.*	*L./min.*	*ml./min./mm. Hg*
R. D.	Rest	7.4	320	6.1	32.4
	Exercise	28.7	1,350	19.7	37.0
	Epinephrine	6.7	324	12.1	31.2
F. D.	Rest	6.3	295	7.5	24.6
	Exercise	20.1	892	14.3	47.9
	Epinephrine	6.0	231	14.8	23.3
S. S.	Rest	7.8	306	6.4	25.1
	Exercise	17.4	669	9.8	34.9
	Atropine and norepinephrine	7.5	342	10.8	20.7
C. R.	Rest	4.7	220	6.7	23.9
	Exercise	13.9	634	10.2	36.5
	Atropine and norepinephrine	6.5	302	13.1	28.1
H. S.	Rest	5.8	263	10.0	22.6
	Exercise	14.2	659	11.3	36.8
	Atropine and norepinephrine	7.1	296	14.2	20.5
F. D.	Rest	6.7	290	9.3	23.5
	Exercise	17.4	696	16.1	45.4
	Atropine and norepinephrine	6.6	288	16.4	24.0
Means	Rest	6.5 ± 1.1	282 ± 35.9	7.7 ± 1.6	25.4 ± 3.6
	Exercise	18.6 ± 5.5	817 ± 277.0	13.6 ± 3.9	39.8 ± 5.5
	Drugs	6.7 ± 0.5	297 ± 38.0	13.6 ± 2.0	24.6 ± 4.2

* Minute ventilation.
† Cardiac output.

TABLE IV

Effect of procedures designed to increase pulmonary vascular pressures on the steady state D_L in six supine subjects

Subject	Condition	M.V.	C.O.	C.V.P.*	D_L
		L./min.	*L./min.*	*mm. Hg*	*ml./min./mm. Hg*
S. S.	Rest	5.3	4.2	3.5	19.6
	Albumin infusion	6.8	9.6	9.6	22.1
	G suit and albumin infusion	4.8	5.8	17.0	19.0
H. S.	Rest	7.4	6.4	−0.3	22.0
	Albumin infusion	6.3	11.7	3.3	23.0
	G suit and albumin infusion	5.4	7.2	8.8	26.0
J. F.	Rest	5.8	6.0	0.5	19.4
	Albumin infusion	4.6	9.7	4.5	21.3
	G suit and albumin infusion	4.8	6.1	16.2	20.2
J. S.	Rest	8.7	10.3	0.5	29.2
	Albumin infusion	8.3	11.6	3.2	29.4
	G suit and albumin infusion	7.2	12.7	7.0	33.4
F. A.	Rest	7.6	7.7	0.9	22.9
	G suit and albumin infusion	7.2	6.3	12.9	21.2
J. B.	Rest	7.1	9.4	8.9	23.6
	G suit and albumin infusion	8.9	10.3	23.0	31.3
Means	Rest (first 4 subjects)	6.8 ± 1.6	6.7 ± 2.6	1.1 ± 1.7	22.6 ± 4.6
	Albumin infusion	6.5 ± 1.5	10.7 ± 1.2	5.2 ± 3.0	24.0 ± 3.7
	Rest (all 6 subjects)	7.0 ± 1.2	7.3 ± 2.3	2.3 ± 3.5	22.8 ± 3.6
	G suit and albumin infusion	6.4 ± 1.6	8.1 ± 2.8	14.2 ± 5.9	25.2 ± 6.1

* Central venous pressure.

TABLE V

Effect of changes in ventilation rate on the steady state D_L in six supine subjects

Subject	Condition	M.V.	C.O.	pCO₂*	(1)† D_L	(2)† D_L	(3)† D_L
		L./min.	L./min.	mm. Hg	ml./min./mm. Hg		
D. G.	Rest	6.5	7.1	38.4	19.9	20.0	19.9
	Air hypervent.	20.8	8.1	24.0	45.7	38.7	49.2
	CO₂ hypervent.	22.5	8.5	42.7	43.2	45.3	42.9
C. W.	Rest	5.0	5.4	38.8	24.5	21.9	22.0
	Air hypervent.	11.7	5.8	27.6	29.1	29.3	29.4
	CO₂ hypervent.	19.5	8.6	46.5	29.8	30.6	29.5
B. S.	Rest	5.4	6.2	40.5	24.6	26.1	25.6
	Air hypervent.	13.9	6.4	29.4	41.4	36.9	43.3
	CO₂ hypervent.	17.8	7.0	46.8	37.1	36.0	39.5
R. R.	Rest	6.8	6.7	41.8	19.9		
	Air hypervent.	12.1	7.3	28.9	27.9		
C. R.	Rest	6.1	9.4	39.3	21.7		
	Air hypervent.	19.4	9.5	24.6	41.2		
R. D.	Rest	6.5	6.8	42.4	23.5		
	Air hypervent.	18.3	6.9	28.6	44.5		
Means	*a) First 3 subjects*						
	Rest	5.6	6.2	39.2	23.0	22.7	22.5
	Air hypervent.	15.5	6.8	27.0	38.7	35.0	40.6
	CO₂ hypervent.	19.9	8.0	45.3	36.7	37.3	37.3
	b) All 6 subjects						
	Rest	6.1 ± 0.9	6.9 ± 1.2	40.2 ± 1.5	22.3 ± 2.2		
	Air hypervent.	16.0 ± 3.6	7.3 ± 1.2	27.2 ± 2.1	38.3 ± 7.1		

* Arterial blood.

† D_L (1) is calculated using arterial blood pCO₂ to determine "r," D_L (2) is calculated using alveolar pCO₂ to determine "r," and D_L (3) is calculated using alveolar pO₂ to determine "r."

effect on D_L was the same. For the reasons already discussed (see Methods), D_L in these three subjects was calculated with "r" determined using arterial blood pCO₂, with "r" determined using alveolar pCO₂, and with "r" determined using alveolar pO₂. The values for D_L calculated by the three different methods are in close agreement.

In summary, voluntary hyperventilation, whether on air or 5.0 per cent CO₂, caused the same apparent increase in steady state D_L as occurred with exercise which induced an equal rise in ventilation rate.

Effect on the steady state D_L of procedures designed to increase pulmonary vascular pressures

A study was made of the effect on steady state D_L of procedures designed to increase pulmonary vascular pressures by pulmonary vascular engorgement. The four subjects who received an intravenous infusion of 5.0 per cent human serum albumin in normal saline (Table IV) had a mean rise in central venous pressure of 4.1 mm. Hg, a concomitant rise in mean cardiac output to 10.7 ± 1.2 L. per minute from 6.7 ± 2.6 L. per minute at rest, and no change in mean ventilation rate. The mean D_L was 22.6 ± 4.6 ml. per minute per mm. Hg in the control period and 24.0 ± 3.7 ml. per minute per mm. Hg with the albumin infusion. In six subjects, inflation of a G suit, in combination with serum albumin infusion, produced a mean increase in central venous pressure of 11.9 mm. Hg without significant change in mean ventilation rate or cardiac output. The mean D_L was 22.8 ± 3.6 ml. per minute per mm. Hg at rest and 25.2 ± 6.1 ml. per minute per mm. Hg with the increased central venous pressure (Table IV). When these 10 instances in which pulmonary vascular engorgement was produced by either albumin infusion alone or G suit inflation with albumin infusion are considered together, a small but statistically significant increase in D_L, amounting to 2.0 ml. per minute per mm. Hg (p < .05) (25), was

TABLE VI

Effect of procedures designed to increase pulmonary vascular pressures on the steady state D_L in three supine subjects

Subject	Condition	M.V.	C.O.	C.V.P.		D_L
		L./min.	*L./min.*	*mm. Hg*		*ml./min./mm. Hg*
B. C.	Rest	7.6	6.1		1.0	24.3
	G suit*	20.8	8.1	up	3.3	43.3
				down	0.7	
	G suit and norepinephrine infusion	14.8	7.2		5.9	29.2
B. M.	Rest	4.9	5.8		−0.4	21.3
	G suit*	8.1	4.2	up	7.3	28.8
				down	1.7	
	G suit and norepinephrine infusion	13.1	5.5		8.8	40.8
J. B.	Rest	7.1	9.4		8.9	23.6
	G suit*	9.4	9.1	up	26.1	28.0
				down	8.9	
Mean	Rest (first 2 subjects)	6.3	6.0		0.3	22.8
	G suit and norepinephrine infusion	14.0	6.4		7.4	35.0
	Rest (3 subjects)	6.5	7.1		3.2	23.1
	G suit*	12.8	7.1	up	12.2	33.4
				down	3.8	

* With alternate application and release of pressure as described in Methods.

associated with the pressure elevation (Table IV). This increase in D_L was not dependent upon an increase in ventilation rate, since ventilation was actually lower in most of the subjects during the period of pulmonary vascular engorgement than during the control period.

In two subjects who had a combination of G suit inflation and norepinephrine infusion, mean CVP was increased by 7.1 mm. Hg without change in the cardiac output (Table VI). The mean ventilation rate, however, also increased. Mean D_L in these subjects increased from 22.8 ml. per minute per mm. Hg at rest to 35.0 ml. per minute per mm. Hg.

The three subjects who were studied during G suit inflation in which the pressure was alternately increased and decreased, as already described, had a significant rise in central venous pressure, but this was also accompanied by an increase in mean ventilation rate. D_L increased from 23.1 ml. per minute per mm. Hg at rest to 33.4 ml. per minute per mm. Hg (Table VI).

In summary, various procedures designed to increase pulmonary capillary pressure by engorging the lungs with blood produced only minor changes in steady state D_L unless they also caused an increase in minute ventilation; D_L showed a substantial increase only when ventilation was increased.

Effect of a combination of albumin infusion with increased ventilation on the steady state D_L

To investigate the possibility that increased pulmonary vascular pressures could enhance the effect of hyperventilation on D_L, three subjects were studied, first at rest, then with hyperventilation, and lastly with hyperventilation during an intravenous infusion of 400 to 500 ml. of 5.0 per cent human serum albumin in normal saline (Table VII). Hyperventilation alone, with a mean ventilation rate of 16.6 L. per minute, increased mean D_L to 37.9 ml. per minute per mm. Hg from 21.7 ml. per minute per mm. Hg at rest. About the same mean ventilation rate, 16.3 L. per minute, accompanied by a 6.8 mm. Hg increase in the central venous pressure produced by the albumin infusion, increased the

TABLE VII

Effect of a combination of albumin infusion with an increased ventilation rate on the steady state D_L in three supine subjects

Condition	M.V.*	C.O.*	C.V.P.*	D_L
	L./min.	*L./min.*	*mm. Hg*	*ml./min./ mm. Hg*
Rest	6.5	7.6	−0.5	21.7
Hyperventilation on air	16.6	7.1	−1.5	37.9
Hyperventilation on air in combination with albumin infusion	16.3	10.3	5.3	39.5

* Mean values for three subjects.

mean D_L to 39.5 ml. per minute per mm. Hg, essentially the same increase as with hyperventilation alone.

In summary, albumin infusions, intended to produce pulmonary vascular engorgement, did not enhance the effect of hyperventilation on D_L, as measured by the steady state method.

B. Studies with D_L determined by the breath holding technique

As shown in Table VIII, 19 supine subjects had a mean D_L of 35.7 ± 8.4 ml. per minute per mm. Hg at rest. Eight of these subjects, with a mean D_L of 38.6 ± 9.7 ml. per minute per mm. Hg supine, had a mean D_L of 34.8 ± 5.9 ml. per minute per mm. Hg when seated (Table IX). This value agrees closely with the mean D_L of 31.7 ± 4.4 ml. per minute per mm. Hg which Ogilvie, Forster, Blakemore and Morton found in eight young male seated subjects (5). Three subjects (ages 10, 10 and 8) were excluded from their group of 11 subjects in determining this mean, in order to make the group comparable to ours in age and size. In the present group of eight subjects, the difference between D_L in the supine and seated positions is not statistically significant.

The procedures required to prepare the subject

TABLE VIII

Breath holding D_L determinations in 19 normal supine subjects at rest

Subject	Age	Surface area	Number of determinations	Mean alveolar volume	Mean D_L
		M.²		*ml.*	*ml./min./ mm. Hg*
P. B.	23	2.10	2	5,570	32.9
H. S.	22	1.72	4	4,450	35.0
B. L.	23	2.14	1	5,570	47.8
C. H.	21	2.14	4	5,090	39.4
Jo. W.	21	2.08	8	5,200	35.7
C. W.	24	1.87	4	3,850	30.0
J. B.	21	2.12	5	4,430	28.0
J. P.	22	2.04	4	5,110	52.4
D. G.	22	2.06	4	5,240	52.2
J. C.	23	1.86	4	3,780	29.8
F. D.	24	2.10	6	6,040	40.9
R. C.	23	2.02	4	6,050	44.2
J. W.	31	1.88	4	5,400	34.6
L. W.	23	2.28	4	4,900	30.1
G. P.	25	2.02	3	5,870	32.6
J. O.	23	1.79	3	4,440	31.1
H. T.	22	2.14	3	5,460	27.2
S. S.	22	1.70	3	3,490	22.8
F. A.	21	1.76	3	3,980	32.4
Mean	23	2.00		4,940±785	35.7±8.4

TABLE IX

Breath holding D_L determinations in eight normal seated subjects at rest

Subject	Age	Surface area	Alveolar volume	Seated D_L	Mean supine D_L*
		M.²	*ml.*	*ml./min./mm. Hg*	
C. H.	21	2.14	4,700	32.2	39.4
Jo. W.	21	2.08	5,730	34.2	35.7
C. W.	24	1.87	4,140	27.9	30.0
J. B.	21	2.12	5,000	32.2	28.0
T. P.	22	2.04	5,380	40.4	52.4
O. G.	22	2.06	6,100	42.5	52.2
J. C.	23	1.86	4,380	27.8	29.8
F. D.	24	2.10	6,400	41.4	40.9
Mean	22	2.03	5,230±815	34.8±5.9	38.6±9.7

* From Table VIII.

for the collection of arterial blood samples and determination of cardiac output produce a small but unavoidable amount of discomfort. To study the effect of this discomfort and accompanying apprehension on the control level of D_L, the D_L was determined in 11 subjects: *a)* before any procedures, *b)* immediately after the arterial needle and venous catheter were put in place, and *c)* after the usual 30 minute waiting period. Mean values were 39.0 ± 8.0, 40.0 ± 10.1 and 38.2 ± 9.9 ml. per minute per mm. Hg, respectively, and are not significantly different.

Effect of exercise and increased cardiac output on the breath holding D_L

D_L and cardiac output were determined in eight subjects while at rest, then during exercise, and finally at rest with cardiac output increased by drugs (Table X). During exercise, mean cardiac output increased to 13.2 ± 2.1 L. per minute from 6.6 ± 1.0 L. per minute at rest while mean D_L increased to 48.9 ± 10.5 ml. per minute per mm. Hg from 39.8 ± 9.7 ml. per minute per mm. Hg at rest. When the cardiac output was increased to 11.9 ± 3.6 L. per minute by drugs, however, the mean D_L did not increase from the resting value.

Four of these subjects also had determinations of cardiac output and D_L during exercise at the same time that their cardiac output was already increased by drugs to determine whether these drugs might have some action which would tend to prevent D_L from increasing (Table XI). The result was not entirely clear-cut. In each sub-

ject, exercise still caused an increase in D_L, although the increase was less than that produced by exercise before the administration of drugs.

The effect of increasing cardiac output by release of arterial tourniquets which had occluded blood flow to the lower extremities for the preceding 15 minutes was also investigated in three

TABLE X

Breath holding D_L, alveolar volume and cardiac output in eight supine subjects at rest, during exercise, and with the cardiac output increased by drugs

Subject	Condition	Alveolar volume	C.O.	D_L
		ml.	*L./min.*	*ml./min./ mm. Hg*
B. L.	Rest	5,570	7.0	47.8
	Exercise	5,990	12.4	57.5
	Drugs*	5,890	14.9	49.6
C. H.	Rest	5.040	6.3	38.0
	Exercise	4,880	13.3	43.4
	Drugs	4,950	9.9	40.7
Jo. W.	Rest	4,750	8.7	33.1
	Exercise	5,270	15.9	47.5
	Drugs	5,410	8.1	33.1
C. W.	Rest	3,760	6.0	28.4
	Exercise	4,100	12.4	36.9
	Drugs	3,500	9.4	27.4
T. P.	Rest	5,100	5.7	52.5
	Exercise	5,460	11.5	64.3
	Drugs	5,160	15.4	45.2
F. D.	Rest	6,080	5.6	39.7
	Exercise	6,330	16.7	48.3
	Drugs	6,150	10.6	42.3
D. G.	Rest	4,780	6.4	50.8
	Exercise	5,330	10.5	58.4
	Drugs	5,030	17.6	53.7
J. C.	Rest	3,930	7.1	28.0
	Exercise	4,450	13.1	34.8
	Drugs	4,060	9.0	29.0
Mean	Rest	4,880±770	6.6±1.0	39.8± 9.7
	Exercise	5,230±740	13.2±2.1	48.9±10.5
	Drugs	5,020±880	11.9±3.6	40.1± 9.5

*Atropine and norepinephrine administration as described in Methods.

subjects. As indicated in Table XII, the mean cardiac output was almost doubled, but there was little change in D_L.

In summary, breath holding D_L was considerably increased, as expected, by exercise which doubled the cardiac output, but doubling the output by drugs or tourniquet release had little effect on D_L.

TABLE XI

Breath holding D_L and cardiac output in four supine subjects at rest, during exercise, with cardiac output increased by drugs, and with exercise while the cardiac output was increased by drugs

Condition	C.O.*	D_L*
	L./min.	*ml./min./mm. Hg*
Rest	6.2 ± 0.7	42.8 ± 11.3
Exercise	13.0 ± 2.7	51.5 ± 12.9
Drugs†	13.2 ± 4.0	42.5 ± 10.6
Exercise with drugs†	16.6 ± 2.6	46.5 ± 9.0

* Means of four subjects.
† Atropine and norepinephrine administration as described in Methods.

Effect of increased ventilation rate on the breath holding D_L

Even though the breath is being held during the determination of D_L, exercise is accompanied by an increase in ventilation rate in the period preceding the breath holding period. The possibility that increased ventilation before the breath holding period could change D_L was studied by having five subjects hyperventilate for a period of five or six minutes immediately prior to the determination of D_L (Table XIII). The mean D_L at rest was 36.4 ± 8.0 ml. per minute per mm. Hg and after hyperventilation was 35.5 ± 7.4 ml. per minute per mm. Hg. Ogilvie, Forster, Blakemore and Morton reported comparable results in a similar study (5).

TABLE XII

Effect of blood pH changes on the breath holding D_L in three supine subjects

Condition	C.O.*	Arterial* pH	D_L*
	L./min.		*ml./min./ mm. Hg*
Rest		7.42	36.3
Arterial tourniquets on the lower extremities	4.4	7.42	33.8
30–40 seconds after the tourniquets were released	8.5	7.35	35.1
After NH_4Cl administration		7.38	35.8
Arterial tourniquets on the lower extremities after NH_4Cl administration		7.38	35.0
30–40 seconds after the tourniquets were released		7.34	37.8

* Means of three subjects.

Effect of increased ventilation rate on the breath holding D_L in five supine subjects

Condition	Alveolar* volume	C.O.*	D_L*
	ml.	L./min.	ml./min./ mm. Hg
Rest	4,460 ± 880	7.0 ± 1.2	36.4 ± 8.0
Hyperventilation†	4,290 ± 820	6.0 ± 4.0	35.5 ± 7.4

* Means of five subjects.
† Hyperventilation for five to six minutes immediately before the breath holding period.

Effect of a combination of increased cardiac output and increased ventilation rate on the breath holding D_L

The breath holding D_L was not changed by either increased ventilation rate prior to the test or increased cardiac output alone. To mimic exercise more closely, two subjects were studied, first at rest, then with cardiac output increased by drugs, and finally with hyperventilation while the cardiac output was increased by drugs. The mean D_L did not change significantly when the mean cardiac output was increased from 6.0 to 10.1 L. per minute by drugs. With the mean cardiac output increased to 10.1 L. per minute, the subjects hyperventilated for five or six minutes immediately prior to the breath holding period, but this also failed to change D_L significantly.

Effect of blood pH changes on the breath holding D_L

Exercise causes a reduction in the blood pH, more in mixed venous than arterial blood. An attempt was made to simulate this change without actual exercise by the use of arterial tourniquets on the thighs and administration of NH_4Cl. Release of arterial tourniquets which have made the legs ischemic for 15 minutes not only increases the cardiac output (18), but also reduces blood pH because a large volume of hypoxic, relatively acidic, blood enters the venous side of the circulation from the previously occluded region (26). To reinforce this drop in pH, subjects were also studied after they were prepared by the oral administration of 20 Gm. of NH_4Cl during the course of the day before the last part of the experiment was carried out. D_L and arterial blood pH, therefore, were determined in three subjects: a) before arterial tourniquets were ap-

plied to the lower extremities, b) while arterial tourniquets occluded blood flow to the lower extremities, c) within 30 to 60 seconds after release of the arterial tourniquets, and d) under all three of the aforementioned conditions after the administration of NH_4Cl. In this study, as in the other studies where pH was determined in conjunction with the breath holding D_L, the arterial blood sample for pH determination was withdrawn from the indwelling arterial needle over the entire 10 second breath holding period. There appears to be no relation between D_L and changes in arterial blood pH brought about by these procedures (Table XII). In addition, a trial was made in two subjects of the effect of maintaining the arterial blood pH above 7.50 by voluntary overbreathing during exercise. This procedure had no apparent effect on the increase in D_L during exercise, as measured by the single breath method.

In summary, exercise causes a reduction in the blood pH, but reduction of the blood pH by experimental methods did not increase the D_L as does exercise. On the other hand, when the reduction in blood pH, usually caused by exercise, was prevented by overventilation, the increase in D_L with exercise was not altered.

Effect of reduction in mixed venous blood oxygen content on the breath holding D_L

Exercise reduces the oxygen content of mixed venous blood without much change in arterial blood oxygen. Conceivably, venous hypoxia could affect the diffusing capacity by changing the resistance of precapillary vessels in the lung and so changing the distribution of blood to the capillaries. To produce venous, but not arterial, hypoxia without exercise, the following procedure was employed. Two subjects breathed 10 per cent oxygen for 10 minutes, lowering the arterial blood oxygen saturation to between 60 and 70 per cent, as estimated by an ear oximeter,[5] and presumably reducing the mixed venous blood oxygen. At the end of this time, for measurement of D_L by the breath holding method, the subject made the usual full expiration followed by a maximal inspiration of the carbon monoxide mixture, which contained 21 per cent oxygen. For the 10 seconds of the measurement, before

[5] Waters Conley.

significant recirculation occurred, this procedure achieved some degree of mixed venous hypoxia with substantially normal arterial blood oxygen saturation. This procedure, tried in two subjects, had a negligible effect on D_L, and increasing the cardiac output with drugs at the same time caused no further change. These results are comparable to the results of a similar experiment reported by Cander and Forster (27).

DISCUSSION

It is well established that exercise increases the pulmonary diffusing capacity. In this study, as in previously reported studies (1–5), exercise caused an increase in D_L, both breath holding and steady state, in all subjects where D_L was determined before and during exercise; and, as others have also observed (5), there was a proportionally greater increase in the steady state D_L than in breath holding D_L. The mechanism of this increase in D_L with exercise was studied by measuring the apparent pulmonary diffusing capacity in situations where one or more of the effects of exercise were reproduced experimentally.

Krogh (1) suggested that change in a stagnant plasma layer in the capillaries could be the mechanism by which exercise increases D_L. Change in the capillary wall permeability or capillary size brought about by lung volume change was suggested by Barcroft (28) as an explanation for the increase in D_L with exercise. Forster (29) has pointed out that it is reasonable to assume that the surface area of the capillary bed increases with exercise, either by dilatation of patent vessels or opening of previously closed ones, as a result of vasomotion or hemodynamic forces. Roughton (12) found that the volume of blood in the pulmonary capillary bed is increased with exercise.

The cardiac output increases during exercise and it has been considered that the increase in D_L is dependent in some way upon this change. This factor was studied by increasing the cardiac output in subjects otherwise at rest and measuring D_L. The cardiac output was increased in groups of subjects by three somewhat different methods: a) drugs, b) albumin infusion, and c) release of arterial tourniquets previously occluding blood flow into the lower extremities. In this study, neither the breath holding nor steady state D_L were increased when the cardiac output was increased by methods other than exercise. In one group (Table III), the mean cardiac output was the same during exercise as when drugs were given, but even though D_L was markedly increased by exercise, D_L did not increase over the resting level when the cardiac output was increased by drugs. In all, 67 simultaneous determinations of cardiac output and steady state D_L were made in 24 subjects under different experimental conditions. There is no significant correlation between the two variables ($r = 0.219$). This is supported by the observation of Johnson, Stein and Kimbel (30) who found that the breath holding D_L of several patients with increased cardiac output due to hyperthyroidism was not significantly different from the D_L found when they had normal cardiac output after they were made euthyroid by therapy.

The effect of increasing pulmonary vascular (pulmonary arterial and capillary) pressure was also investigated. Increased pulmonary vascular pressure is probably not the cause of an increased D_L with exercise since D_L increases significantly with moderate exercise, whereas in normal man there is little or no increase in pulmonary arterial pressure with moderate exercise (31, 32). This study was made, however, to determine whether an increase in D_L could be produced by procedures designed to increase pulmonary vascular pressures. With experiments in which the ventilation rate was substantially unchanged, increase in pulmonary vascular pressures, as reflected by a fairly large increase in the mean central venous pressure, was accompanied by only a small increase in the steady state D_L. Lewis, Forster and Beckman (23) did not find a significant increase in the breath holding D_L with inflation of a G suit.

Increases in ventilation rate, however, whether or not produced by exercise, were uniformly found to be associated with increases in steady state D_L. Hyperventilation alone, without any change in the cardiac output, was accompanied by an increase in steady state D_L in all subjects studied, whether they hyperventilated while breathing air or 5 per cent CO_2. The increase in steady state D_L with hyperventilation in one group of subjects (Table V) was about the same

as the increase produced by exercise in another group of subjects (Table III) where the increase in mean ventilation rate was nearly the same. There is a highly significant correlation ($r = 0.787$) between steady state D_L and ventilation rate in the 67 determinations on 24 subjects mentioned above in whom the correlation between D_L and cardiac output was not significant. Taken alone, these data suggest that the effect of ventilation rate on steady state D_L is sufficient to account for the increase in D_L which occurs with exercise. Increases in breath holding D_L, however, also occur with exercise when the rate of ventilation cannot be a factor during the test period itself because the breath is being held. Hyperventilation before the breath holding period, to mimic the course of ventilation during exercise, was without effect on D_L. Similar results have been reported previously (5). It would appear that the increase in breath holding D_L which occurs with exercise depends upon some mechanism other than increase in cardiac output or ventilation rate.

An attempt was made to enhance the effect of hyperventilation on D_L by simultaneously increasing cardiac output or by producing pulmonary vascular engorgement. The increase in steady state D_L produced by hyperventilation, however, was not enhanced by simultaneous pulmonary vascular engorgement. In addition, the breath holding D_L, which was not increased by hyperventilation alone before the test period, likewise failed to increase when hyperventilation was combined with increase in cardiac output caused by drugs.

Exercise reduces pH and pO_2 and increases pCO_2 of mixed venous blood. Conceivably, such changes could affect the precapillary vascular system of the lungs in such a way as to alter the distribution of blood and increase D_L. For this reason, the effect on the breath holding D_L of changes in pH and gas content of mixed venous blood was studied in small groups of subjects. Reducing pH did not cause an increase in breath holding D_L and, likewise, partially counteracting the acidosis of exercise by hyperventilation did not prevent an increase in D_L from occurring with exercise. Forster and co-workers (13, 29) found only a slight increase in breath holding D_L when 6.0 per cent CO_2 was included in the in-

spired mixture, but a more significant increase in breath holding D_L of 20 per cent in nine subjects who had breathed 7.5 per cent CO_2 for 10 minutes. Observations were also made on the effect of reducing mixed venous pO_2. The breath holding D_L was neither increased by the procedure which was designed to reduce the mixed venous pO_2 while allowing a normal arterial pO_2, nor by combination of this procedure with an increased cardiac output. The procedure which probably produced changes in the mixed venous blood most like those of exercise was the release of tourniquets from ischemic lower extremities, which increases cardiac output and releases blood with a reduced pH and pO_2 and an increased pCO_2 into the venous side of the circulation. This, however, failed to increase D_L as measured by the breath holding method. Breath holding D_L was determined about 30 to 40 seconds after release of tourniquets, and it is possible that the changes caused by this procedure were not present for sufficient time to increase D_L.

In summary, the breath holding D_L was not increased significantly by any procedure except exercise, while the steady state D_L was increased by hyperventilation alone to about the extent that it was increased by exercise with a similar ventilation rate.

There are other factors which are known to be important in determining the actual value of D_L, but are not applicable in this study. The values of D_L reported here are for apparent pulmonary diffusing capacity of the lung as a whole, which includes the factors of resistance to passive diffusion of carbon monoxide through the "membrane" and resistance to reaction with hemoglobin. Roughton, Forster and co-workers (12, 33–36) have measured the reaction rate of CO with hemoglobin and devised a method for determining separately the diffusing capacity of the membrane (D_m). While the reaction rate of CO with hemoglobin must be determined in order to measure D_m, the apparent D_L can be measured without consideration for kinetics of the CO+Hb reaction, though it is obviously playing a part in the actual value obtained. The reaction rate of CO+Hb would have been applicable in this study only if it were being considered as a possible mechanism by which exercise increases D_L or if it were being changed by procedures used

to mimic the effects of exercise. There is no indication that a significant change in reaction rate was produced in this way. Reaction rate can be affected by changes in mean pO_2 of pulmonary capillary blood (37), but the present experimental procedures probably altered this little. In a normal subject breathing 21 per cent oxygen, the oxygen tension of pulmonary capillary blood probably increases so rapidly when the blood is exposed to alveolar gas tensions (38), that the mean capillary pO_2 is not greatly altered by moderate changes in the pO_2 of pulmonary arterial blood. Even if the mean capillary pO_2 were calculated as the arithmetic mean of pulmonary arterial and systemic arterial blood pO_2 the present experimental procedures would have produced little change. From the data in Table III, it can be calculated that the mean A–V oxygen difference was 3.7 volumes per cent for subjects at rest, 6.0 during exercise, and 2.2 when the cardiac output was increased by drugs. For normal subjects these values would correspond to pulmonary arterial pO_2 values of approximately 48, 40 and 60 mm. Hg, respectively, (pH 7.30). The greatest difference in mean capillary pO_2 between these groups should have been less than 10 mm. Hg, and the effect of this on the reaction rate of carbon monoxide with hemoglobin is negligible for present purposes (37).

Steady state D_L determinations are influenced by the mixed venous COHb concentration (11, 12) and to determine D_L as accurately as possible, this must be taken into account. The present study has been concerned primarily with factors which change D_L rather than with the absolute value of D_L. For the reasons already given, therefore, "back pressure" does not seem to be significant in the interpretation of the results of this study and no correction was made for capillary pCO. In the case of the breath holding D_L, the effect of capillary pCO has also been found to be almost insignificant at near normal alveolar O_2 tensions (11, 39).

The answer to the question of why D_L increases with exercise is incomplete. By these methods, changes in cardiac output and pulmonary vascular pressures are without significant effect. With the steady state method, ventilation rate is of great importance and in the use of this method for investigative purposes, con-

sideration must be given to the effect of changes in ventilation rate. The mechanism by which increased ventilation rate causes an increase in steady state D_L has not been determined. During exercise or with hyperventilation, the mean volume of the lung is somewhat increased and it has been suggested in the past that increase in lung volume is the mechanism by which exercise causes an increase in D_L (28). Grape and Tyler (40) found that the steady state D_L increased when the alveolar volume was increased, but the amount of rise they observed is not sufficient to account for the rise which occurs with exercise. Increased ventilation rate may possibly increase the steady state D_L by causing an increase in the perfused area of the pulmonary capillary bed or by causing a more even ventilation-perfusion ratio throughout the lung (29). The data available from this study do not bear on these possibilities.

SUMMARY

1. A study has been made of the means by which exercise increases the pulmonary diffusing capacity for carbon monoxide (D_L) in normal subjects, using both the steady state and breath holding techniques.

2. Increases in cardiac output produced by means other than exercise caused no significant change in D_L as measured by either the steady state or breath holding technique.

3. Steady state D_L was found to be very sensitive to the ventilation rate, and by hyperventilation alone, D_L was increased as much as by exercise at the same minute ventilation. Breath holding D_L was not affected by hyperventilation preceding the measurement.

4. Although the effect of increased ventilation rate could explain the increase in steady state D_L with exercise, an adequate explanation for the increase in breath holding D_L which occurs during exercise was not obtained.

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