ON THE FLUCTUATION IN THE COMPOSITION OF THE ALVEOLAR AIR DURING THE RESPIRATORY CYCLE IN MUSCULAR EXERCISE.

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INTRODUCTION.

THE measurement of the circulation rate in man has been attempted by various methods, none of which has come into general use. The problem is important in physiology, and possibly more so in medicine, where physiological explanations are sought for the phenomena of disease. For that reason, among others, the recently devised ethyl iodide method of measuring the circulation rate published by Yandell Henderson(5) in 1925, has attracted considerable attention. In this method the subject breathes a certain concentration of ethyl iodide vapour for a given time, during which the amount of ethyl iodide taken up by his blood and the concentration of the vapour present in his alveolar air are determined; from these results, given a knowledge of the partition coefficient of ethyl iodide between blood and air, the circulation rate can be calculated. In such an experiment, the reliability of the method of sampling alveolar air is obviously of fundamental importance. Yandell Henderson uses an automatic modification of Krogh's Copenhagen method, whereby in successive expirations small samples are taken from the last portions of air expired, and it is assumed that the composition of the mixed sample obtained in this way is the same as the composition of alveolar air. Alveolar air, however, varies in composition during the respiratory cycle; in expiration its CO_2 increases and its oxygen decreases, while the opposite changes take place in inspiration. For calculation of the circulation rate it is necessary to know the average alveolar composition throughout the experimental period, which comprises many respiratory cycles. How far the composition of Yandell Henderson's sample approximates to that average is not clear. The fractions of his sample are withdrawn from the alveoli at the same phase of successive respiratory cycles; that phase may coincide with the moment at which the fluctuating composition of the alveolar air is passing through its mean, but it cannot be proved to do so until the extent of the fluctuation is accurately known. It may be that in some types of breathing (*e.g.* at rest) the coincidence is good, while in others (*e.g.* during work) there is a discrepancy. The aim of the work to be described in this paper was to obtain some definite information about the degree to which the composition of alveolar air varies throughout the respiratory cycle, and if possible to compare its average composition with that of the last portion of the expired air which constitutes Yandell Henderson's alveolar sample. All the observations were made during muscular exercise, because the amount of variation in the alveolar concentrations, and the discrepancy (if any) between their average values and those of the Yandell Henderson sample, are both likely to be greater when the breathing is deep and the respiratory exchange increased.

It would be very difficult to follow the changes in the alveolar air by a direct method, such as taking Haldane-Priestley samples at different phases of the respiratory cycle, because every sampling would disturb the rhythm of the breathing. An indirect method, however, is applicable during expiration, for if the variation in the composition of the air *passing out of the mouth* in a single expiration is observed, it is possible to deduce what changes must have occurred in the alveolar air to give rise to the observed changes in the expired air. This was the method adopted. For observing the changes in the expired air some form of apparatus was required which would receive a single expired breath, and divide it up into a number of separate successive portions suitable for measurement and analysis. This, moreover, must be done without offering any obstruction to the expiration, and without altering its rate or its volume. Such an apparatus was devised, based on a system of automatically operated valves which directed successive portions of an expired breath into six small rubber bags. It was made of a size large enough to accommodate breaths of 1½ to 3½ litres, which might be given by a subject doing moderate muscular work. In the following pages the apparatus and the experiments carried out with it will be described first. Then the results will be given, together with the deductions that may be drawn from them, including certain conclusions as to the size and the nature of the respiratory dead space.

DESCRIPTION OF APPARATUS.

The apparatus consists of the following parts:

I. A Central Chamber, through which the subject breathes; in its walls are six openings, fitted with electrically operated valves, leading into six small rubber bags.

II. A System of Switches which operate the valves automatically.

III. A Recording System, which simultaneously registers the action of the valves and the breathing of the subject on a revolving drum.

IV. A Douglas Bag.

These will now be described in detail.

I. *The Central Chamber* is an oblong air-tight brass box which is shown in longitudinal section in Fig. 1. When the subject breathes through the mouthpiece *M*, air enters at *A* through a Rosslyn valve fixed by adhesive into a hole in the brass, and passes out at *B* through another similar valve and a piece of corrugated rubber tubing into the Douglas bag. Three of the six openings in the sides of the chamber are shown on the left hand side of the figure, leading into short lengths of

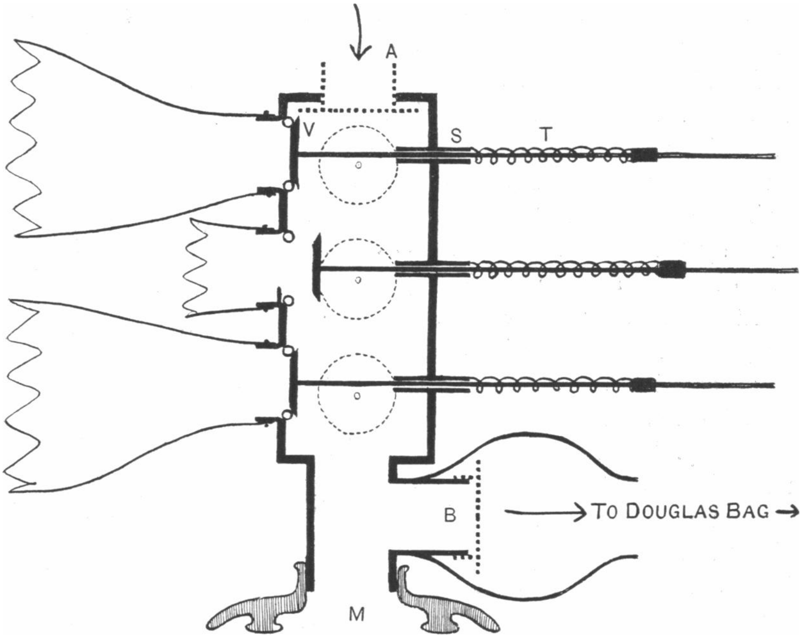

Fig. 1. Diagrammatic longitudinal section of the central chamber of the apparatus.

brass tubing to which are fitted the necks of collapsible thin rubber bags of about 500 c.c. capacity. Each opening is provided with a valve *V*, made of a disc of metal, bevelled at the edge, and carried on the end of a thin metal rod. The valve seating is a ring of narrow rubber tubing fixed with adhesive to the margins of the opening in the brass wall; when the valve is pressed on to it, wet or dry, it makes an air-tight contact; the rubber must be kept free from grease. The rod occupying the disc passes through a close-fitting and well-greased brass sleeve, *S*, soldered into the opposite side of the chamber. Its other end is attached to an electromagnet which opens the valve (the middle one of the three valves is shown open), while a spring *T* keeps the valve shut when the magnet is not in action. These three valves and their sleeves occupy two opposite sides of the chamber, which is square in cross section; the other two sides are similarly occupied by the other three valves (shown as dotted circles in the figure) and their sleeves.

Fig. 2 is a photograph showing the chamber with its six rubber bags, together with the magnets and their connections, mounted in a wooden frame. Each bag has a narrow rubber side-tube leading through a glass four-way-piece to three gas-sampling bottles of the kind described by McCann and Hannan and figured by Clark-Kennedy and Owen(1). Three of them are sufficient to hold the contents of one bag; they contain a mixture of equal volumes of glycerine and saturated sodium chloride solution, over which expired air can be kept for 6 hours without loss of CO_2. The magnets are operated by a 240-volt current from the mains passing through a resistance of 120 ohms.

II. *The System of Switches.* Each rubber bag is associated with a double mercury switch, seen in Fig. 2 and shown diagrammatically in Fig. 3. Attached loosely to the surface of the bag is a hinged aluminium rod *R*, carrying a transverse ebonite arm with two steel pins dipping into ebonite cups of mercury. When the bag is collapsed, contact is made in the cup *M*, which is in the circuit of the electromagnet controlling the valve of the same bag. When the bag is filled with air it takes up

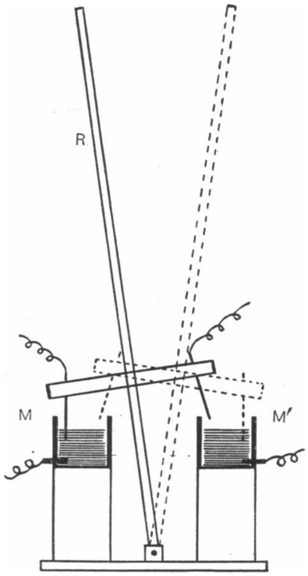

Fig. 3. Diagram of a mercury switch.

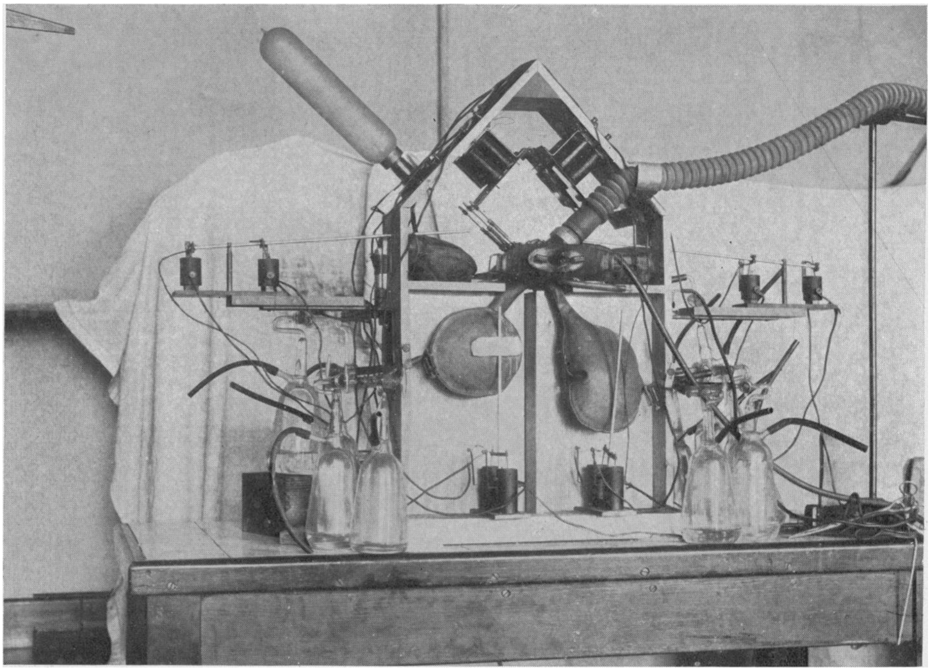

Fig. 2. Photograph of the apparatus showing the central chamber, electromagnets, mercury switches, and small rubber bags, mounted in a wooden frame, together with the sampling bottles in position.

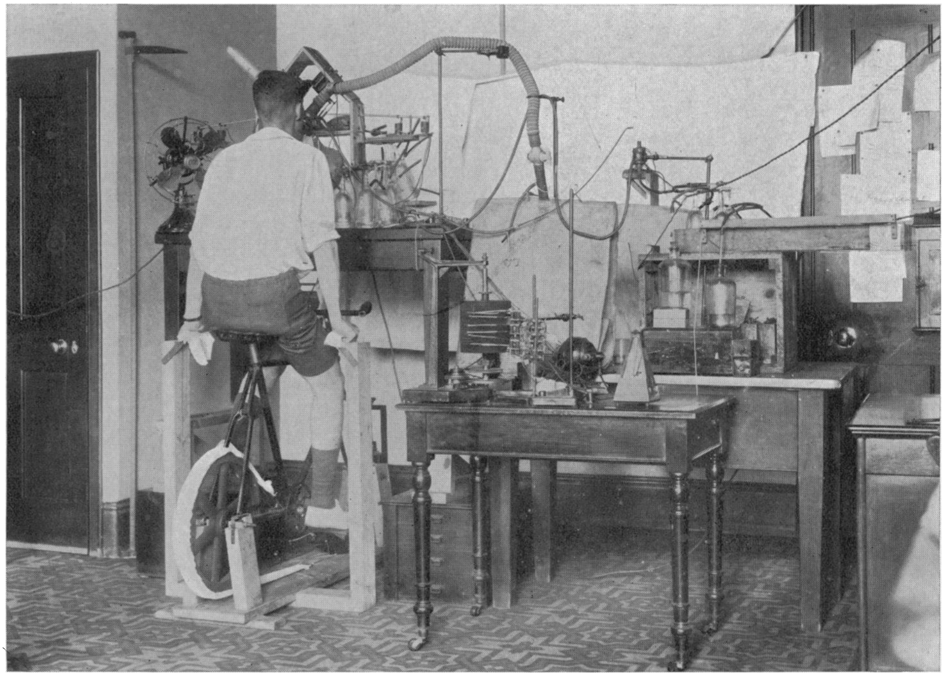

Fig. 4. Photograph showing the arrangement of the whole apparatus for an experiment.

the position shown by the dotted lines: contact is broken at *M*, whereupon the valve of this bag closes; contact is made at *M′* in the other mercury cup which is in the circuit of the magnet that opens the valve of the next bag in the series. In this way each bag as it becomes full closes its own valve and opens that of its neighbour. In an experiment, this process is begun by switching the main current into the circuit of the first electromagnet, and if air is blown into the central chamber it then continues automatically down the series of six bags until the filling of the last one short-circuits the current.

III. *The Recording System.* In the circuit of each magnet is an ordinary signal-marker, which records on a smoked drum the time during which that magnet was in action. A pressure record of the breathing is also taken on the drum by means of a polygraph tambour connected through fine rubber tubing with the inside of the central chamber; this is very sensitive and gives an accurate record of the time occupied by inspiration and expiration respectively. A Jacquet clock marks fifths of seconds.

IV. *The Douglas Bag* holds up to 70 litres of air. It is connected with the expiration valve of the central chamber through corrugated rubber tubing and a 3-way tap, so that the expired air may either be collected or be allowed to escape as desired.

DESCRIPTION OF EXPERIMENTS.

The apparatus was designed to accommodate the rapid and deep breathing of muscular exercise, and care was taken that no air-way should be less than 0·45 sq. in. in cross section. At the same time the dead space of the valves, which is the whole capacity of the central chamber, was kept as small as possible, and for this purpose its superfluous corners were filled with sealing-wax; this instrumental dead-space is 160 c.c. If an expired breath is to be collected in the six bags of the apparatus, it is obvious that at the beginning of expiration these 160 c.c. will contain fresh air, which, during expiration will be swept with the expired air into the bags. In order to find out how quickly this instrumental dead space is washed out, air containing a known uniform concentration of CO_2 was blown into the apparatus from a Douglas bag. In this as in all subsequent experiments the valves furthest from the mouthpiece were the first to open, so that the dead space might be washed out as quickly as possible. The air in the Douglas bag and in the small bags was then analysed, and in one experiment gave the following figures:

Douglas bag	...	4·18 p.c. CO_2
1st bag	...	2·66 ,,
2nd bag	...	4·09 ,,
3rd bag	...	4·16 ,,
4th bag	...	4·17 ,,
5th bag	...	4·18 ,,
6th bag	...	4·17 ,,

A second experiment gave a similar result. The figures show that the washing out is completed by the time the second bag is full.

After this preliminary observation, two sets of experiments were carried out on two subjects. In all these the object was to obtain a breath during muscular exercise, divide it into 5 or 6 successive portions, and determine the changes in gaseous composition from one to another. Six experiments were performed on the subject J.K.M. and eleven on the subject R.S.A. The procedure was essentially the same in them all though there were some minor variations in the first set. The following are the details of a typical experiment.

The subject sat erect on a bicycle ergometer, and the apparatus was so placed that he could take the mouthpiece in his mouth without discomfort or hindrance to his breathing. The general arrangement is shown in Fig. 4. He inspired from the atmosphere through the central chamber, and expired again to air through the 3-way tap on the tube leading to the Douglas bag. As soon as he was comfortable he began to ride the bicycle, at 72 turns of the pedals per minute, keeping time with a metronome. The load on the ergometer was constant for a given experiment, but was varied in different experiments in order to give different volumes of tidal air. The work was continued for 10 minutes, and a fan was used to keep the subject cool in the second half of that time. If a person riding an ergometer under these conditions pays no attention to his breathing, he involuntarily breathes in time with his pedalling, taking one breath to every two, three, or four turns of the pedals, according to the severity of the work, and probably changing at intervals from one ratio to another if his respirations become uncomfortably deep or shallow. In these experiments such a sudden change during a critical period was to be avoided; so the ratio of breathing rate to pedalling rate was decided on beforehand, and the subject consciously kept to it. Both the rate of breathing and the rate of pedalling were so chosen (after numerous trials) as not to involve excessive over- or under-breathing; a minor degree of one or the other was likely to occur, but would not prejudice the results so long as the breathing during the time of collection of expired air was uniform.

Exactly at the end of the ninth minute of work the 3-way tap of the Douglas bag was turned, and from then till the end of the tenth minute the expired air was collected in the Douglas bag, while the respirations were counted. About half-way through the tenth minute, during an inspiration, the expired air tube was suddenly clamped just beyond its valve, and at the same moment the current to the apparatus was switched on. This caused the valve of the first small rubber bag to open, so that the succeeding expiration passed first into this bag and then into the other five in succession as the automatically operated valves came into action[1]. During the following inspiration the current was switched off, and the clamp on the expiration tube released, allowing the expired air during the rest of the minute to pass into the Douglas bag as before. By this whole procedure a single expiration, in the middle of the tenth minute, was diverted in six distinct and successive portions, into the six small bags. This breath will be called the "divided expiration." Meanwhile an assistant had started and stopped the rapidly revolving drum of the recording system at such times as to give a record of the operation of the valves and the duration of the divided expiration. At the end of the tenth minute, the tap of the Douglas bag was closed, and the subject ceased work.

Immediately after that the contents of the six small bags were quickly sucked into the sampling bottles; it had been found out beforehand that if this was carried out inside 2 or 3 minutes, the absorption of CO_2 by the rubber walls of the bags was negligible. The expired air from the six small bags, and the expired air in the Douglas bag, were then measured and analysed. Measuring in the case of the small samples was done in a 700 c.c. burette over glycerine and salt mixture, and the samples were returned to the sampling bottles for analysis. From the air in the Douglas bag two samples were taken for analysis, and the rest measured in a spirometer. Analyses for oxygen and CO_2 were done in duplicate on two Henderson-Haldane burettes; when agreement was not satisfactory they were repeated.

The drum records were measured and from them were calculated the times of opening and closing of the six valves, and the time of beginning

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[1] At the end of the expiration the sixth bag (which was rather larger than the others) might be only partly full, in which case its valve might remain open, but no air could escape from it again because a one-way rubber spear-valve was fitted just inside its neck.

of the next inspiration, taking the beginning of the divided expiration as zero.

The gas volumes, measured moist at room temperature, were recalculated for 37° C., moist. The oxygen intake, CO_2 output, and respiratory quotient were calculated for each bag separately, for the whole divided breath, and for the expired air of the whole tenth minute.

Finally the CO_2 curve for the divided breath was plotted by the method illustrated in Fig. 5. The whole volume of the breath was set out along the abscissa, and subdivided into six parts corresponding to the volumes of the six portions of the breath in the small bags, the first portion to be expired being placed on the left; on each of these subdivisions as base a rectangle was drawn, with its height representing the concentration of CO_2 in the corresponding bag. The area of each rectangle (volume × CO_2 concentration) represents the actual amount of CO_2 in the corresponding bag, while the combined areas of the six rectangles will be equivalent to the total amount of CO_2 in the whole breath. The upper limit of the combined area is a "stepped" curve, which represents literally the concentrations found in the six portions;

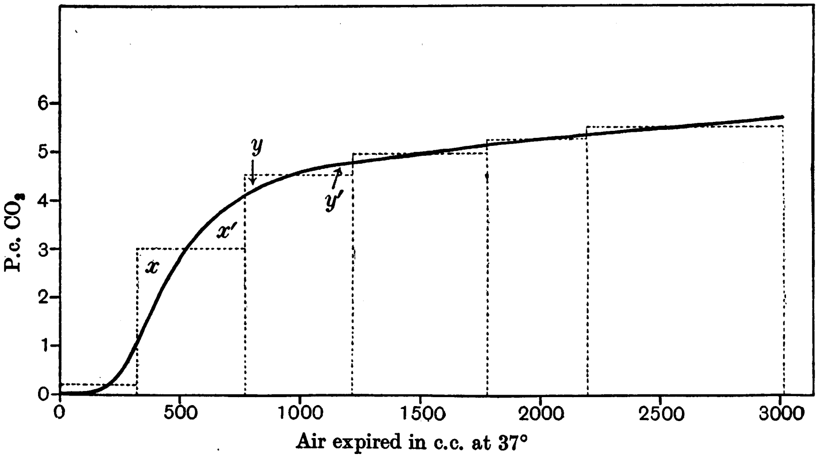

Fig. 5. Diagram showing the method of plotting the CO_2 curve of an expired breath (Exp. 12).

Bag No.	Volume in c.c.	P.c. CO_2
1	324	·22
2	444	2·97
3	450	4·52
4	562	4·98
5	417	5·27
6	829	5·53

however, the CO_2 concentration in the actual air expired does not change in this intermittent stepped fashion but continuously, and should be represented by a smooth curve. If this smooth curve is to be an accurate representation, it is necessary that the area of the part of it corresponding to each bag shall be equal to the area of the rectangle corresponding to that bag, since each of these areas is equivalent to the amount of CO_2 in the bag. To achieve this in practice, the smooth curve was first drawn "by eye" through the tops of the rectangles (plotted on ruled paper) and the areas were compared by counting squares in the figures x and x', y and y', etc.; if necessary, the curve was then altered until $x = x'$, $y = y'$, and so on. In this way, by trial and error, a reasonably smooth curve was arrived at, which satisfied the condition stated above. In every experiment there was found to be very little latitude in the placing of this curve—the six rectangles fixed it remarkably closely. Curves drawn in this way are more accurate than smooth curves drawn merely through the mid-points of the tops of the rectangles.

Eleven experiments of this kind were performed on the subject R.S.A. and six similar ones in the earlier set on the subject J.K.M. Three experiments were discarded on account of irregularities in the breathing of the subject or in the operation of the automatic switches. There remain five on J.K.M. and nine on R.S.A. The results of these will now be given and discussed.

RESULTS AND DISCUSSION.

I. *The curve of* CO_2 *concentration in an expired breath.*

The smooth CO_2 curves obtained in the manner just described in the fourteen experiments under discussion, are shown in Fig. 6, and the general data of the experiments are given in Table I. The curves represent the results of all the experiments done that were not demonstrably defective; they are all reproduced because on their combined evidence rests the principal conclusion of this paper, namely, that the CO_2 curve of the expiration given in these experiments by a subject doing moderate muscular work on an ergometer has a typical shape. Beginning at zero, every curve exhibits an **S**-shaped rise in the CO_2 concentration up to about 1000 or 1200 c.c.; thereafter, in nine cases it proceeds as a straight line, and in the remainder its deviation from a straight line is so small that it can be ascribed to slight irregularities in breathing, or to minor experimental defects. Furthermore, this straight part of the curve, in every case but one, is not horizontal but slopes gently upwards. The

TABLE I. General data of Experiments 1 to 14.

						Last minute*			
Exp. No.	Subject	Duration mins.	Ergometer tension lbs.	Rate of pedalling per min.	Rate of breathing per min.	CO_2 output c.c. at 37°	O_2 intake c.c. at 37°	Tidal air c.c. at 37°	Volume divided breath c.c. at 37°
1	J.K.M.	5	6	80	20	1549*	1628*	3195	2447
2	J.K.M.	5	5	80	20	1480*	1428*	3030	2830
3	J.K.M.	5	4	88	22	1140*	1208*	2430	2501
4	J.K.M.	5	4·5	88	22	1050*	1091*	2100	2293
5	J.K.M.	8	4·5	88	22	2433	2418	2228	2679
6	R.S.A.	10	6	72	36	2343	2282	1890	2038
7	R.S.A.	10	6	72	36	2358	2147	1940	1967
8	R.S.A.	10	6	72	36	2234	2051	1931	2079
9	R.S.A.	10	6	72	18	2117	2062	2880	3285
10	R.S.A.	10	6	72	18	1696	1702	2370	3103
11	R.S.A.	10	6	72	18	1771	1770	2356	2665
12	R.S.A.	10	8	72	18	2105	2148	2760	3026
13	R.S.A.	10	8	72	18	2652	2233	3520	3123
14	R.S.A.	10	9	72	18	2597	2470	3410	3260

* Last *half* minute in Nos. 1, 2, 3 and 4.

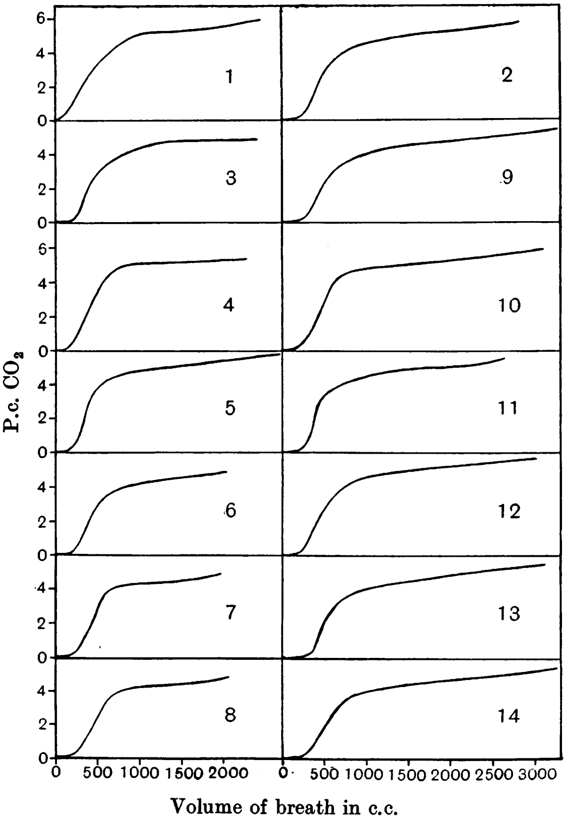

Fig. 6. The CO_2 curves obtained in Experiments 1 to 14.

typical curve, therefore, is made up of two parts: the first rises from zero with a double (**S**-shaped) inflection, and is continued into the second, which is a straight line sloping slightly upwards. (Fig. 5 shows a good example on a larger scale.)

The explanation of this constant shape of the curve seems fairly clear. The whole curve represents the changing CO_2 concentration in the air that is entering the small bags of the apparatus, some of which has come up from the pulmonary alveoli (alveolar air), and some from the bronchi, the trachea, the mouth, and the central chamber of the apparatus, that is, from the dead space of the subject and of the machine (dead space air). The **S**-shape of the first part is due to the mixing of alveolar air rich in CO_2 with dead space air that contains little or no CO_2. The alveolar air tends to come up in axial streams, moving more rapidly in the axes of the respiratory tubes, and more slowly next their walls where it is delayed by friction; hence the CO_2 appears in the small bags of the apparatus before the dead space is completely "washed out," and the curve obtained is not rectangular but doubly inflected. The effect of this dilution of alveolar air with fresh air is however a progressively diminishing one; it explains the **S**-shape of the first part of the curve, but it cannot account for the slope of the second straight part, which is a uniform one. That uniform upward slope in the second part means that *the air which left the alveoli* contained a steadily increasing percentage of CO_2 as expiration progressed, for, when the dead space has once been washed out, air will pass from the alveoli to the small bags of the apparatus almost unchanged; admittedly the axial stream phenomenon still operates, but the sliding of one portion of air past another, which this involves, can have little effect on the slope of the curve when that slope is not great. The same argument applies to such eddies as may occur in the chamber of the apparatus, and it has already been seen that about 800 c.c. of air (contents of first two bags) are sufficient completely to wash out this instrumental dead space. Finally, if it be granted that the air in the alveoli is uniformly mixed, then the straight part of the experimental curve represents the CO_2 concentration in alveolar air, and it slopes up because in the second part of expiration that concentration is increasing at a uniform rate.

II. *The physiological dead space in deep breathing.*

An attempt will be made later to define the physiological factors which influence the slope of the alveolar CO_2 curve during the whole of expiration. So far the actual curve has been obtained directly in the latter half or two-thirds of expiration, and it shows that whatever those factors are, they act together during that time in a uniform manner to produce a uniform upward slope. It seems likely, then, that they will also act together in the same uniform manner during the earlier part of expiration, when the experimental curve is not identical with the alveolar air curve, but is modified by the admixture of the dead space air. If that be so, the alveolar air curve for the earlier part of expiration can be reconstructed by prolonging the straight part of the experimental curve to the left, as has been done by the dotted line *BD* in Fig. 7. The line *DC* now represents the CO_2 concentration of all the air that left the alveoli and also reached the small bags of the apparatus. That air is smaller in volume than the total expired air collected in the bags, the difference being the volume of the dead space, but it must contain the same amount of CO_2 as the total expired air, because the dead space air contains practically none. Since amount of CO_2 is represented in Fig. 7 by area (volume of air × CO_2 concentration), this fact enables the point *D* to be fixed, by the simple procedure of making the area of the figure *FDCG* (representing amount of CO_2 in the air from alveoli) equal to that of the figure *OABCG* (representing amount of CO_2 in expired air). When that is done the distance *OF* represents the total dead space, instrumental plus physiological. This reconstruction, then, affords

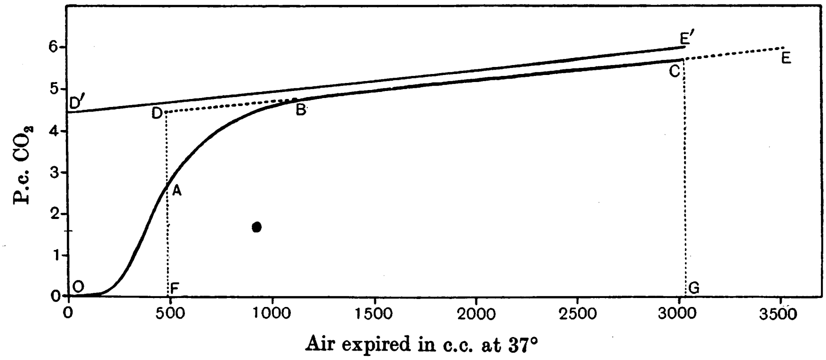

Fig. 7. Diagram to show the method of deducing the alveolar CO_2 curve and the dead space from the experimental CO_2 curve (Exp. 12).

The flat part *BC* of the experimental curve *OABC* is prolonged to the left to *D* until the area *FDCG* equals the area *OABCG*, *DF* and *CG* being perpendicular to *OG*. *OF* then represents the volume of the dead space. *DC* is prolonged to the right to *E*, over a distance equal to *OF* (when measured horizontally). The whole line *DE* then represents the changing concentration of alveolar CO_2 throughout expiration.

a method for determining the physiological dead space in deep breathing during work.

The physiological dead spaces have been determined by this method from all the curves shown in Fig. 6. In practice it was found easier to make the areas equal by algebraical calculation instead of actually counting squares; the dead space was regarded as containing ·03 p.c. CO_2.

In the five experiments on the subject J.K.M. the physiological dead space was found to vary from 228 c.c. to 417 c.c. (measured moist, at 37° C., and prevailing barometric pressure), but it bore no definite relation to the volume of the tidal air. Those experiments, as the curves show, were in other respects more irregular and less satisfactory than the subsequent ones on the subject R.S.A. In the latter (Nos. 6 to 14), the dead space varied from 283 c.c. to 392 c.c., and bore a very definite relation to the volume of the tidal air[1], as will be seen in Table II and Fig. 8. As the size of the breath increases, the dead space increases proportionally over the range covered by the experiments, *i.e.* from 1890 c.c. of tidal air to about 3½ litres. It is not clear, however, whether the rate of breathing has a separate influence on the dead space, over and above that of the depth, although it is suggestive that the three points 6, 7 and 8 are a little out of the line established by the remainder; 6, 7 and 8 are the experiments where the breathing was 36 per minute, while in the rest it was 18 per minute.

TABLE II. Dead space and tidal air in Experiments 6 to 14.

Exp. no.	Average tidal air in tenth minute c.c. at 37°	Physiological dead space c.c. at 37°
6	1890	283
8	1930	290
7	1940	280
11	2356	287
10	2371	295
12	2760	327
9	2880	361
14	3410	378
13	3520	392

These results may be compared on the one hand with those of Haldane and Douglas, and on the other with those of Krogh and

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[1] "Tidal air" means average tidal air for the last minute of the experiment. The whole volume of the divided breath sometimes differed from this (see Table I), but as the difference was due to the divided expiration having been cut short too soon or unduly prolonged, rather than to an irregularity in the inspiration that preceded it, it is more reasonable to seek a relationship between the dead space and the tidal air than between the dead space and the volume of the divided breath.

Lindhard. Haldane and Douglas(3) calculated the CO_2 dead space in Douglas both at rest and at various speeds of walking, determining the

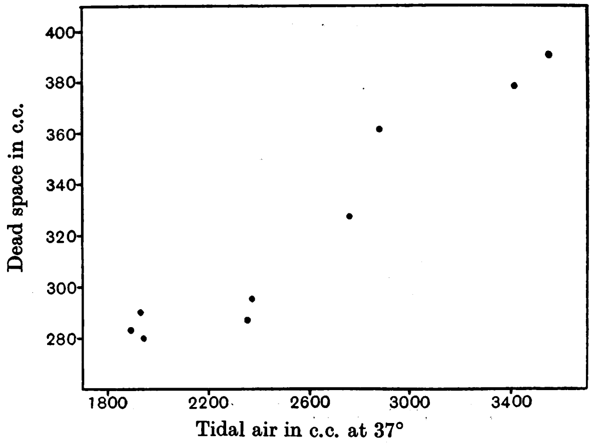

Fig. 8. The relation of dead space to tidal air in Experiments 6 to 14.

average tidal air, the CO_2 in the expired air, and the CO_2 in the alveolar air. The alveolar air was obtained by Haldane and Priestley's method, taking the mean of inspiratory and expiratory samples. The resulting dead space was 213 c.c. (moist, at 37° C. and prevailing barometric pressure) at rest for a tidal air of 457 c.c., and at the various levels of work it increased very regularly with the increasing tidal air up to 675 c.c. for a tidal air of 3145 c.c., when the subject was breathing 60·9 litres of air and consuming 2543 c.c. of oxygen (at N.T.P.) per minute. Our figures in the first place are considerably lower than these; *e.g.* in Exp. 14 when the ventilation was 63·2 litres and the oxygen consumption 2047 c.c. per minute, the dead space was only 391 c.c. for a tidal air of 3410 c.c. Further, while our figures confirm the conclusion of Douglas and Haldane that the dead space does increase with increasing depth of breathing, they indicate a much less rapid increase (about 8 c.c. per 100 c.c. tidal air) than do Douglas and Haldane's figures (about 19 c.c. per 100 c.c. tidal air). These two discrepancies appear to be due to one cause, which was pointed out by Krogh and Lindhard(6) and recognised by Douglas(2), namely, that the Haldane-Priestley method of sampling alveolar air is not reliable when the CO_2 output is increased by muscular work. It takes time to make the deep expiration involved, and during that time CO_2 can pass from the rapidly circulating blood in the lungs into the rapidly diminishing volume of air in the alveoli, in sufficient quantities to raise the CO_2 concentration in the alveolar air well above what it is at the beginning or at the end (as the case may be) of an ordinary expiration. Hence, in exercise, the Haldane-Priestley alveolar air contains more CO_2 than average alveolar air, and the dead space calculated from it is too high. As the amount of work increases, the CO_2 output increases, while the time taken to give the Haldane-Priestley sample remains the same; hence the error becomes greater, and the dead space appears to rise with increasing tidal air more rapidly than is really the case.

Krogh and Lindhard(7), on the other hand, maintain that the dead space at rest is of the order of 100 c.c. and that with deep breathing (at rest also) its maximum rise is to about 200 c.c. They based this conclusion on the results they obtained with Siebeck's hydrogen method, or modifications of it; their views on the dead space at rest are discussed fully by Haldane(4). They also give, however, the results of a few experiments not unlike those described in this paper, and performed during work. Their subject expired into a recording spirometer, which, by operating electric contacts, caused two or three samples of air to be taken from between the subject's teeth into evacuated mercury sampling tubes, in the course of one expiration. A third or fourth sample was taken of the very end of the breath, from the expiratory tube, just beyond the valve. The samples were analysed, and the corresponding volumes obtained from the spirometer record. Four experiments are mentioned; three of them have three samples each, the fourth has four and its results are plotted, CO_2 p.c. in sample against volume. All four indicate that during the latter part of the breath, the CO_2 p.c. rises along a straight line; the slope of the line is steeper than in our experiments, but this was probably because the breathing was much slower. The fourth is the only one in which the CO_2 percentage in the expired air is given, and is therefore the only one in which the dead space can be even approximately calculated by the reconstruction method given above. In that experiment, using the line which Krogh and Lindhard have drawn through their four somewhat irregular points, it works out at something in the neighbourhood of 100 c.c. This is in accord with their theory, based on the hydrogen experiments, that the dead space in deep breathing is not greatly increased. It is, however, a single observation (or only very weakly supported by the other three), and a less complete one than any single one of the experiments in this paper. Moreover, the respiratory quotients for the four samples in this experiment are ·64, ·65, ·66, ·61, values which seem surprisingly low for a man doing muscular work, and absorbing 1830 c.c. oxygen per minute. It is possible that, at the time of this breath, the subject, who had been giving Haldane-Priestley samples very shortly before, was not in respiratory and circulatory equilibrium. However that may be, it seems clear from our more extensive application of a method similar to that of Krogh and Lindhard, that the dead space in breathing during moderate exercise is of the order of 300 c.c. to 400 c.c. when the tidal air is between 2 litres and 3½ litres. While these results differ quantitatively from Haldane's, they do not conflict with his picture of what happens in atria and air-spaces during deep breathing, and they support his contention that the dead space should be regarded as a convenient physiological conception rather than a specified anatomical space.

III. *The curve of alveolar* CO_2 *concentration during expiration.*

So long as CO_2 is passing from the blood in the lung capillaries into the alveolar air, the alveolar CO_2 concentration during expiration must rise, but it is not easy to analyse accurately the factors that determine the rate at which that rise will occur. There seem to be two quantities, variations in which are of the greatest importance; one of them changes in such a way as to favour a decreasing rate of rise, the other an increasing rate. They are:

1. The difference in CO_2 pressure between the blood in the capillaries of the lung and the alveolar air. This is the force which makes CO_2 diffuse across the endothelium and the alveolar wall. During expiration this force is diminishing, because
   (*a*) the CO_2 pressure in the alveoli is rising while that in the blood is steady, or falling;
   (*b*) the oxygen pressure in the alveoli is falling; hence hæmoglobin is being less quickly oxidised, and is less effective in keeping the CO_2 pressure of the blood high by displacing CO_2 from combination with alkali.

This diminishing pressure difference means a diminishing influx of CO_2 into the alveoli. This factor acting alone (assuming for the moment a constant alveolar volume) tends to cause the alveolar CO_2 to rise, but to rise less and less rapidly, during expiration; if it were the only factor the curve of alveolar CO_2 concentration would slope upwards, but less and less steeply, *i.e.* it would be concave downwards.

2. The volume of alveolar air (or, strictly, of air-sac air) into which a given amount of CO_2 diffuses. As this diminishes, the effect of the added CO_2 in raising the CO_2 concentration increases. This factor acting alone (assuming for the moment a constant pressure difference and a constant

influx of CO_2) would cause the alveolar CO_2 to rise, and to rise more and more rapidly, during expiration; if it were the only factor, the curve would slope upwards, more and more steeply, *i.e.* it would be concave upwards. (It may be objected that a constant pressure difference, as assumed, does not involve a constant CO_2 influx, because as the air-sac shrinks the area of its walls, across which diffusion takes place, also diminishes; this, however, does not destroy the argument, because the volume must always diminish more rapidly than the area—therefore the increasingly upward trend of the curve remains.)

Such theoretical considerations, then, make it appear likely that two factors are principally responsible for the rise in alveolar CO_2 during expiration, namely the continued diffusion of CO_2 into the alveoli, and the progressive diminution in alveolar volume, the former tending to accelerate the rate of rise and the latter to retard it. How far the two factors, acting together, will counterbalance each other cannot be foretold on theoretical grounds, but from direct experimental observation of the alveolar CO_2 curve in the latter two-thirds of moderately slow expiration during work, it seems that their combined effect is to make the alveolar CO_2 rise at a uniform rate.

Provided the expiration is delivered at a uniform rate and not in a jerky or irregular fashion, these factors may be expected to act in a regular manner, and to counterbalance each other, not only in the part of expiration where the alveolar CO_2 curve has been obtained directly, but throughout the whole of expiration. Hence it was assumed that the alveolar CO_2 curve for the earlier part of expiration could be reconstructed by a backward prolongation of the straight part of the experimental curve (Fig. 7). A similar procedure also can be applied to the right-hand end of the experimental curve. The point *C* represents the last of the air that left the alveoli and also succeeded in reaching the small bags of the apparatus. A further quantity of air left the alveoli, but failed to reach the small bags; at the end of expiration it occupied the dead space. This further quantity can be represented by prolonging the experimental curve to the right over a distance equal to the dead space as already determined; this is done in Fig. 7 by the dotted line *CE*. The whole line *DE* now represents the CO_2 concentration in air leaving the alveoli, and therefore in alveolar air, throughout expiration. In the figure, the line *DE* has been re-drawn as *D'E'*, with *D'* above zero on the abscissa: it can then be said that while air entering the small bags of the apparatus is changing its CO_2 concentration in the way represented by the experimental curve *OABC*, the air in the alveoli is doing so along the line *D'E'*.

In this example, *D'E'* rises from 4·5 p.c. CO_2 at the beginning of a 3-litre expiration occupying 1·77 seconds, to 6·02 p.c. CO_2 at the end, a rise of 1·52 p.c. or ·51 p.c. per litre. In most of the other curves, the slope is similar. In the nine experiments on the subject R.S.A. it varies from ·46 p.c. CO_2 to ·59 p.c. CO_2 per litre of air expired, and it is not appreciably different in the shorter breaths from what it is in the longer ones. In moderate work, therefore, under the conditions of these experiments, the fluctuation of alveolar CO_2 concentration throughout the respiratory cycle is of the order of ·5 p.c. CO_2 per litre of tidal air.

In the last paragraph but one, it was stipulated that the expiration be delivered at a uniform rate; the argument in that paragraph, indeed the whole reconstruction of the alveolar CO_2 curve, rests on the assumption that it was delivered uniformly, and it was in order to justify this assumption that the records of the times of opening and closing of the valves were taken on the revolving drum. Those obtained in Exp. 12, which was a typical one, are shown in Fig. 9. The straight lines in the lower part of the figure indicate the time during which each bag was open, and are drawn from the measurements made on the tracing. They show a certain amount of overlap between adjacent bags, *e.g.* Bag 2 remained open for several hundredths of a second after Bag 3 opened, but this was inevitable, for had the overlap been reduced too far a stage would have been reached at which the expired air would have encountered a series of brief obstructions. In order to

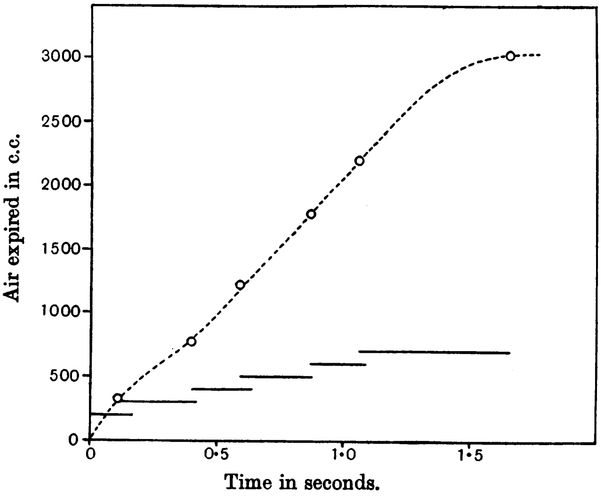

Fig. 9. Diagram showing the rate at which the divided breath was expired (Exp. 12).

The straight lines indicate the times during which the six valves were open. They correspond to Bags 1 to 6, in order from left to right.

The curve is obtained by plotting the volume of air already collected when each bag opens, against the time that has elapsed since the expiration began. The highest point corresponds to the closure of the sixth bag, very shortly after which the next inspiration began.

plot the curve (in Fig. 9) showing the rate at which air was expired, it was assumed that each bag ceased to fill with air at the moment when, as shown by the record, its successor opened; this may involve a slight error, but not more than two or three hundredths of a second. The curve indicates that, on the whole, the expiration was delivered at a uniform speed. There is, however, a small temporary hurry at the beginning, which appears only in some of the records, and may be instrumental; and there is a definite slowing down at the end of expiration, which is a feature of all records, and is a physiological phenomenon. In this kind of breathing, expiration slows off a little towards the end, and there may even be a slight pause before inspiration begins.

The time records therefore show that the assumption mentioned is in the main justifiable, but it is well to enquire how great may be the effect of the irregularities that do occur, especially on the mean value of the alveolar CO_2 curve during expiration, which is to be discussed later on. The initial hurry is slight; it makes no difference to the flat part of the experimental curve, or to the slope of the alveolar air curve; it does make the dead space, as calculated, a few c.c. too large. The final slowing, however, needs further consideration. It means that if the experimental curve were plotted against *time* along the abscissa instead of against *volume*, it would be continued further to the right, and would bend downwards a little towards the end, thus becoming more nearly horizontal. Its average height would in that case be a little higher than that of the curve plotted against volume, and the same applies to the reconstructed alveolar air curve. At the same time, the prolongation to the right that represents the air in the dead space at the end of expiration would now be a continuation, not of the main slope of the curve, but of the more nearly horizontal part, and that would make the average height of the reconstructed alveolar air curve a little lower than it would otherwise have been. The average value, therefore, of the alveolar curve should not be seriously in error because of the slowing at the end of expiration, and the dead space calculation will not be affected by it.

IV. *The respiratory quotients of successive portions of an expired breath.*

The respiratory quotients of the successive portions of the breaths obtained in the experiments on the subject R.S.A. are given in Table III.

TABLE III. Respiratory quotients, Experiments 6 to 14.

Experiment no.	Bag 1	Bag 2	Bag 3	Bag 4	Bag 5	Bag 6	Whole divided breath	Expired air in tenth minute
6	1·77	1·03	1·01	1·00	1·01	—	1·02	1·03
7	1·60	1·12	1·11	1·09	1·08	—	1·11	1·10
8	1·26	1·08	1·10	1·09	1·07	—	1·09	1·09
9	1·28	1·06	1·06	1·07	1·05	1·03	1·06	1·03
10	1·19	1·00	1·03	1·01	1·01	·99	1·01	1·00
11	2·00	1·01	1·01	1·02	1·01	·99	1·01	1·00
12	1·90	·97	1·01	1·00	1·00	·97	·99	·98
13	2·23	1·26	1·32	1·27	1·25	1·24	1·28	1·19
14	1·12	1·04	1·09	1·07	1·06	1·04	1·06	1·05

In every case the first portion of the breath has an R.Q. much above the average for the whole breath. The remaining four or five portions have respiratory quotients that are close to the average; if anything, there is a slight tendency for the R.Q. to fall as the breath proceeds, and that of the last portion is always a little below the average.

The respiratory quotients for whole breaths which are in the neighbourhood of 1·00 may be looked on as normal for the tenth minute of such work as was performed. Those which are higher are probably due to overbreathing, but since the respiratory quotients for the whole tenth minute in those cases were also high, the overbreathing was not confined to the divided breath, and a certain steady degree of overbreathing in the tenth minute does not in any way impair the value of the results for the purposes of this paper.

The value for the R.Q. in the first portion is subject to a large experimental error, because the CO_2 concentration and the reduction in the oxygen concentration are both small; but this cannot account for the uniformly high figures obtained. They are explained by the fact that the air which is last inspired and first expired comes in contact with a large area of mucous membrane in the respiratory passages, and also, in all probability, with some alveoli in the respiratory bronchioles, alveolar ducts, etc., as distinct from the air-sacs proper. This air has a very low CO_2 pressure; therefore CO_2 diffuses into it from the mucous surfaces and alveoli with which it comes into contact very much faster than it would into alveolar air containing 4 or 5 p.c. of CO_2. On the other hand, while this air has a higher oxygen pressure than alveolar air, it does not lose its oxygen much more readily to the blood in those surfaces and alveoli, because above the alveolar oxygen pressure the oxygen dissociation curve of hæmoglobin is relatively flat. The net result is that its gain of CO_2 is out of proportion to its loss of oxygen, and its R.Q. is therefore high. The remaining portions are not affected in this way; their respiratory quotients are fairly steady, and therefore close to the average value for the whole breath.

The existence of the initial high R.Q. arising in this way makes it clear that the R.Q. of alveolar air and the R.Q. of mixed expired air cannot be the same. Mixed expired air contains alveolar air and air from the dead space: if the latter has an R.Q. higher than the R.Q. of the mixture, then the former, the alveolar air, must have a lower R.Q. than the mixture. How much lower depends on the relative proportions of alveolar and dead space air, or in other words the ratio of dead space to tidal air. In deep breathing, the dead space is small compared with the tidal air; hence, as the figures in Table III show, the alveolar R.Q. is only slightly lower than the expired air R.Q. On the other hand, in quiet breathing at rest, the dead space is a much larger fraction of the tidal air, and the alveolar R.Q. may fall considerably below the expired air R.Q., as Haldane first pointed out. Pearce(8), however, argued that if the alveolar air does not prove to have the same R.Q. as the expired air for the same period, then the method of sampling of alveolar air is at fault; these considerations make it clear that Pearce's criterion is unsound.

This phenomenon also emphasises the difference between the anatomical dead space and the physiological, functional or effective dead space. The diffusion of CO_2 referred to above takes place into the anatomical dead space, which is the actual space in the respiratory passages from the mouth down to some undefined boundary that separates it from alveolar air. The physiological dead space is a conception involving the actual volume of the respiratory passages, the rate and volume of ventilation, the rate of diffusion of oxygen and CO_2, and various other factors. It is best defined as that volume of atmospheric air which must be added to the air that leaves the alveoli to make its concentration of oxygen or of CO_2 the same as that of the expired air. On account of the diffusion of CO_2 into the respiratory passages, the physiological dead space for CO_2 must always be smaller than that for oxygen.

V. *Automatic sampling of alveolar air during exercise; average alveolar air.*

Several automatic devices have been introduced for the purpose of collecting alveolar air by withdrawing the last few c.c.'s of successive breaths throughout an experiment. Yandell Henderson(5) used one such device in his ethyl iodide method of determining the circulation rate, and considered the alveolar air so obtained to be equivalent to average alveolar air. Later Clark-Kennedy and Owen(1) published records of "alveolar" air obtained during exercise by a modified sampler, operated electrically; they did not consider that their sample had the composition of average alveolar air, but they did claim that its CO_2 concentration could not be lower, nor its oxygen concentration higher, than those of average alveolar air.

The curve in Fig. 7 may be used to assess the value of these methods. Had one of them been used in this experiment, it would have given a sample containing about 5·75 p.c. CO_2, corresponding to the extreme end (*C*) of the experimental curve. The mean value for this whole curve is 5·26 p.c., so in this case the automatic sample would have contained about ·5 p.c. more CO_2 than the average alveolar air during expiration. In most of the other experiments the discrepancy would be of the same order, because the curves have much the same slope; it would of course be smaller in the shorter curves.

[Clark-Kennedy and Owen's apparatus was actually used in Exps. 13

and 14, and the samples obtained contained 5·28 and 5·35 p.c. CO_2 respectively; the corresponding values read off from the curves were 5·54 and 5·43 p.c. The curves of course represent single breaths while the samples represent the mean of 18 breaths, and cannot be expected to agree exactly with the curves.]

It remains uncertain whether the average CO_2 concentration of the alveolar air during expiration is the same as the average concentration throughout the whole respiratory cycle. Direct evidence seems unobtainable; theoretical considerations rest largely on assumptions. At first sight it appears that in inspiration, after a slight delay due to the re-inspiration of CO_2-laden air from the dead space, the alveolar CO_2 concentration will fall at a steady rate, because it is influenced by the same factors as co-operated to produce the steady expiratory rise, only acting in the opposite direction. But there is one important difference; in expiration the alveolar volume is diminished by abstraction of air of the same composition as that left behind; in inspiration, the alveolar volume is increased by introduction, not of air similar in composition to that already there, but of fresh air containing no CO_2. This speeds up the fall in CO_2 concentration in the first half of inspiration, so that in all probability the average alveolar CO_2 concentration during inspiration is lower than that during expiration. If that be so, the average alveolar CO_2 concentration for the whole cycle is also lower than the average during expiration, and the error involved in regarding an automatic sample as average alveolar air is greater than that already indicated. It is safe to conclude that, in this type of breathing, the error involved by regarding the CO_2 concentration of an automatically collected sample as that of average alveolar air is at least + 10 p.c.; this error will re-appear in any calculation of circulation rate (such as Yandell Henderson's) which so regards the automatic sample, and will probably be as great for ethyl iodide as it is for CO_2.

(For a similar reason it is clearly unsafe to assume that the CO_2 given off into the alveoli during expiration is half of that given off during the whole cycle; had that assumption been permissible it would have been possible from the data in Fig. 7 and Table I to calculate the subject's residual and reserve air.)

SUMMARY.

1. An apparatus has been devised for collecting a single expired breath from a human subject during moderate muscular exercise, and dividing it into six separate successive portions suitable for measurement and analysis.

2. The curve representing the varying concentration of CO_2 in the successive portions of an expired breath in moderate muscular exercise is shown to have a typical shape, which is described and illustrated.

3. From this curve the varying concentration of CO_2 in the alveolar air during expiration can be deduced graphically; the alveolar CO_2 rises steadily during expiration at the rate of about ·5 p.c. per litre of air expired.

4. From this curve also the size of the physiological dead space of the subject can be deduced; it is found to lie between 300 c.c. and 400 c.c., increasing slightly with increase in the volume of the tidal air.

5. In this type of breathing, a sample obtained from the last air expired by Yandell Henderson's method of sampling alveolar air (or by similar methods), will have a CO_2 concentration at least 10 p.c. higher than the average concentration of CO_2 in the alveolar air throughout the respiratory cycle.

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