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HYPOXIA AND HYPEROXIA IN MAN

Immediate effects on ventilation and heart rate

A Thesis

presented to

The University of Glasgow

for the

Degree of Doctor of Philosophy

in the

Faculty of Medicine

by

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Lammas, 1969.

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FOREWORD

Shortage and excess of oxygen are not everyday occurrences, though normal men may meet them in unusual environments, and abnormal men through deficiency of gas exchange and its treatment.

When relatively gross alterations in inspired oxygen concentration are experimentally imposed, the responses observed may be relevant to the understanding and care of men in these abnormal situations; but they may also illuminate the mechanisms of adjustment to minor changes which are part of normal homeostasis:

> "..... the rapid adjustments meeting the fleeting emergencies of the day, resting primarily on a dual gaseous exchange, are dependent mainly upon pulmonary ventilation and the circulation of the blood..... Perfectly useless, the one without the other, they work in closest cooperation through the aid of similar and common mechanisms of control .... Stimulation of the chemoreceptors produces simultaneous effects on ventilation and circulation, but the manner in which this multiple action is accomplished is not yet known .... a common basic reaction of all peripherally lying chemoreceptors to chemical changes could meet in a simple way the rigid requirements of coordination of ventilation and circulation."
>
> Gesell 1939.

The manner of this coordination is still somewhat of a mystery, but it appears not to be met in such a basically simple way.

This thesis reports a study of ventilatory and cardiovascular responses in man: it attempts to identify the chemoreflex component of these responses by studying their timing and sequence during changing states immediately following alterations in the composition of inspired gas. This approach owes much to Dejours:

> "... Le principe de Claude Bernard .... implique que les effets d'une cause perturbante sont, d'une part maximum, d'autre part relativement simples et univoques au début de la phase déséquilibre, avant que des réactions secondaires, dont l'integration réalise le nouvel état d'équilibre, ne viennent obscurir la 'singularité de la perturbation initiale."
>
> Dejours 1967.

A general introduction summarises the present state of knowledge and theory about chemical factors in ventilatory control, since this is relevant to the whole thesis. Subsequently each section is introduced by an additional review appropriate to its subject.

The contents page following gives only the main headings in each section: the first page of each section gives a more detailed plan of contents.

ACKNOWLEDGMENTS

Professor R.C. Garry, as Head of the Department and Supervisor, has always been a source of helpful advice and encouragement.

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The students, technicians, patients and others who acted as subjects submitted cooperatively to unrewarded tedium.

More generally, I have had from time to time the benefit of discussion, criticism, encouragement and help with statistics, variously from several colleagues; it is appropriate to mention in some or all of these contexts, Drs. J.V.G.A. Durnin, J.F. Lamb, and T.D.M. Roberts, and again Professor Garry.

Mrs. E. Stirling brought the necessary good-humoured forbearance to the task of typing the script.

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SYMBOLS AND ABBREVIATIONS

V	Gas volume	T	tidal
$\dot{V}$	Ventilation rate; gas volume per unit time in the sense used by convention in respiratory physiology (more strictly $\int\dot{V}$)	I	inspired
		E	expired
		A	alveolar
f	Frequency of breathing, breaths per min		
R	Respiratory exchange ratio		
P	Partial pressure or tension in:	I	inspired gas
		E	expired gas
		A	alveolar gas
		a	arterial blood
		$\bar{v}$	mixed venous blood
		jv	jugular venous blood
h	Frequency of heart beat, beats per min		
BP	arterial blood pressure		
CBF	cerebral blood flow		
PAP	pulmonary artery pressure		

GENERAL INTRODUCTORY REVIEW

Chemical Factors in ventilatory control

Medullary chemoreceptors

Arterial chemoreceptors

Stimuli

Recent advances

(1) Oscillating stimuli

(2) Rapidity of response

(3) Neural control

Methods of study in experimental animal

Ventilatory chemoreflex function in man

Chemodenervation

CO_2 responsiveness at different O_2 levels

Timed breath-by-breath studies following changes in inspired gas

Approach to present experiments

On leaving CO_2 uncontrolled

Relationship between cardiovascular and ventilatory changes

Present experiments

CHEMICAL FACTORS IN VENTILATORY CONTROL

A great deal of attention has been given to this subject in recent years. There have been a series of international symposia directly devoted to it, or inevitably turning upon it. The field will be briefly scanned as a background to the present work.

Theories of chemical regulation have alternated between the simple, inclusive, single factor theories, and the various ways of describing an admittedly complex system. Haldane & Priestley's assertion in 1905 that ventilation was regulated in such a way as to keep alveolar PCO_2 constant could be said to have stood the test of time, with certain qualifications: Gesell (up to 1939) and Winterstein (up to 1961) repeatedly upheld acidity as the ultimate stimulus and they, also, are well served in current postulates about central regulation. Gray (1950) developed a multiple factor theory summating the several stimuli. Lloyd and Cunningham pursued a different analysis embodying new concepts of interaction as well as mathematical sophistication (1963). Neilsen (1936, 1951), meanwhile, championed CO_2 as the ultimate

factor. It can be said that regulation is effected by complex interacting negative feedback systems for arterial PCO_2, PO_2 and pH, such that the claim of a change in any one of them to act as an effective ventilatory stimulus is dependent on the levels of each of the others. And chemical regulation is itself superimposed on a complex background of neural adjustment.

The control system is served by two sets of receptors, central and peripheral, and their function continues to be elucidated.

Medullary chemoreceptors

Recent research points to cerebral extracellular pH, which varies very readily with changes in arterial PCO_2 and in cerebral blood flow (CBF), as the essential determinant of the background level of ventilation. The action is widely believed now to be upon the medullary surface chemorecptors (Mitchell, Loeschcke, Massion & Severinghaus, 1963). This supercedes the view that the neurones of the respiratory centre were themselves stimulated by a rise in PCO_2. (However, the claim for these surface receptors as the essential region of central chemosensitivity is not uncontested: Cozine & Ngai (1967) believe them to be only one relatively small component

of central sensitivity). Should there be a persistent increase in other ventilatory stimuli - such as increased peripheral chemoreceptor traffic in hypoxia - the braking effect of the secondary CSF alkalinity is gradually diminished as CSF pH is adjusted back towards normal by movement of bicarbonate ions (Mitchell, 1965). Thus despite a persistent reduction in blood and CSF PCO_2, elevated ventilation can be maintained, because the CSF pH is restored to normal. The situation is complicated by the simultaneous effects of the respiratory gas tensions on the cerebral blood flow.

It is generally accepted, since the review and additional evidence presented by Watt, Dumke & Comroe (1943), that the direct effect of hypoxia on the respiratory centre is depression. (These authors did also say, however, that in chemodenervated dogs, hypoxic depression was reversed after the first few minutes and ventilation sometimes rose above normal).

The arterial chemoreceptors

These have been recognised since 1927 (Heymans & Heymans). Structural descriptions were the life's work

of De Castro (up to 1967), and functional studies are chiefly associated with the names of Heymans, Neil & Joels, Comroe & Schmidt, Daly, and now more recently with Torrance & Lee, Purves & Biscoe.

For practical reasons, the carotid bodies have been studied far more extensively than the aortic and the term chemoreceptor is frequently used as synonymous with carotid bodies. As the aortic body contribution to ventilatory responses is relatively small (Comroe, 1939), this is in the present context perhaps justified.

Stimuli: The chemoreceptors have been shown to be stimulated by:

i) hypoxaemia - reduced P_aO_2 (Von Euler, Liljestrand & Zottermann, 1939)

ii) hypercapnia - increased P_aCO_2, and decreased pH, independently of each other, but only when the oxygen tension is normal or low.(Joels & Neil, 1961)

iii) decreased blood flow, implying decreased oxygen flow: yet they are not stimulated by reduced arterial oxygen content, with normal blood flow. Decreased blood flow will even stimulate them in hyperoxic conditions. Stimulation by adrenaline and noradrenaline is related to this effect (Landgren & Neil, 1951).

iv) various drugs. (Comroe, 1964)

More recently the role of these receptors in normal physiological control has received more serious attention: they have come into the limelight as fine-adjustment controls rather than last-ditch defenders against the unlikely hazard of severe hypoxia. This swing of emphasis is associated with the following observations and hypotheses:

1) Greater response to oscillating stimuli

Yamamoto & Edwards suggested in 1960 that altered amplitude of an oscillation in CO_2 level in the arterial blood might give rise to altered stimulation of chemoreceptors despite a constant mean P_aCO_2 value. Support for this has been found experimentally (in cats) by Purves (1965, 1966). This mechanism would allow of increased stimulation in exercise, despite normal mean blood levels. Attempts to alter artificially the amplitude of oscillations in man have so far produced no supportive evidence for the theory (Cunningham, Elliott, Lloyd, Miller & Young, 1965).

2) Rapidity of response

Stimulation of the afferent chemoreceptor fibres, or changes in perfusing blood, will modify the current or the next respiratory cycle (Black, McCloskey & Torrance, 1966; Dutton, Hodson, Davies & Chernick, 1967).

3) Neural control

The chemoreceptors have an efferent autonomic nerve supply which influences their activity (Floyd & Neil, 1952; Eyzaguirre & Lewin, 1961). Neural control may affect not only the blood flow but also the oxygen consumption of the glomus cells, thus varying the relationship between the arterial PO_2 and the tissue PO_2 (Biscoe & Purves, 1965, 1967a,b,c). Such control would make it quite possible for chemoreflex activity to increase at the start of exercise even before there is any change in level or pattern of the arterial blood gases (Neil, 1966).

Information about ventilatory chemoreceptor activity in experimental animals has been obtained by:

1. Application of stimuli to chemoreceptors

i) via the lungs or venous blood

ii) directly in the arterial supply of the chemoreceptor by close arterial injection, or by perfusion of the isolated carotid bifurcation region (and more recently of the artificially separated aortic body region).

Measurement of response

i) by measuring ventilation

ii) by recording from few- or single-fibre preparations of the afferent nerve.

2. Observation of the effects of various stimuli before and after chemoreceptor denervation:

The most detailed information about the chemoreceptors themselves has been obtained from stimulating them directly and measuring impulse traffic in their afferent fibres. Knowledge of their function in the response of the whole animal must take account of whether or not ventilatory change reflects chemoreceptor input: the respiratory centre may deal with increased chemoreceptor impulses in different ways according to whether or not the whole animal is subjected to the same change as the chemoreceptors. Simultaneous measurement of ventilation and impulse traffic in response to stimuli applied to the

whole animal is likely to give the most useful information for an integrated picture. Such studies include those by Loeschke's group, and Leitner's, and they seem to indicate a close relationship between chemoreceptor activity and ventilatory response in the particular circumstances studied (Loeschke, Mitchell, Katsaros, Perkins & Konig, 1963; Leitner, Pages, Puccinelli & Dejours, 1965).

Ventilatory chemoreflex function in man

There are certain rare clinical situations in which the opportunity has presented itself to study human subjects before and after destruction of carotid body function:

1) After removal of the carotid bodies, bilaterally, as a treatment (unsuccessful) for intransigent asthma: Holton & Wood (1965) reported findings in two such cases.

> The subjects showed loss of the ventilatory response to hypoxia after operation. Ventilation was depressed when 10% oxygen was inspired 2 weeks after operation. One recovered some response later. Both retained a response of heart rate. Resting ventilation remained at a lower level than before operation. The complete absence of response, at least initially, indicated the lack of any important ventilatory function attributable to the aortic chemoreceptors.

2) After bilateral removal of carotid body tumours. Wright has studied such a case (personal communication to Holton & Wood) and one is reported in this thesis. The response to hypoxia was lost in both cases.

3) After bilateral carotid endarterectomy. Wade and co-workers have reported studies on 4 such cases, and found a diminished response to CO_2 at given levels of hypoxia, and a fall in blood pressure during hypoxia where previously there was a rise. (Wade, Larson, Hickey & Ehrenfeld, 1969).

Apart from these fortuitous opportunities, the knowledge of chemoreceptor function in man is largely indirect: normal subjects are studied by altering inspired gas composition, and measuring alveolar or arterial blood gas levels as the stimuli, and ventilation as the response; the overall response has then to be interpretated in terms of peripheral and central effects and their interactions. Alternatively, since separation of peripheral from central chemoreceptor activity cannot be carried out anatomically in man, the other possibility is to separate them temporally. This has been the basis of several studies.

Response to carbon dioxide at different oxygen levels

The study of the overall response of ventilation to different combinations of CO_2 and oxygen, in combination with other variables, has yielded a great deal of information and it has been possible to quantify responsiveness in man (Neilsen & Smith, 1951; Lloyd, Jukes & Cunningham, 1958; Lloyd & Cunningham, 1963). This method relates the total response to the differently combined stimuli, but it does not help to elucidate their several sites of action: because peripheral chemoreceptors respond both to hypoxia and to CO_2, the total response to a certain combination of the two, minus that to the same PCO_2 in oxygen, will measure the hypoxic component, but not the peripheral chemoreceptor component. Also the counteracting effect of hypoxic depression in the medulla detracts from the total response, although an accompanying rise of CO_2 minimises the cerebral hypoxia by increasing blood flow. So many variables determine the total responsiveness that it is not surprising that variations are considerable, between individuals, and from one occasion to another (Schaefer, 1958; Jennett, 1968). The conclusions which have been made from studies of CO_2

responses may be summarised thus:

1) There is a linear response of ventilation to an increasing P_ACO_2 (or P_aCO_2) in the range from normal up to about 60 torr; below this range the slope of the response curve is probably less steep. If jugular venous blood PCO_2, or that in the bag-lung-blood complex during rebreathing CO_2 in oxygen, is plotted as the abscissa, a more completely linear relationship is demonstrable, suggesting parallel changes between the PCO_2 measured, and the actual central chemoreceptor stimulation (Lambertsen, Kough, Cooper, Emmel, Loeschcke & Schmidt, 1953; Landmesser, Cobb, Peck & Converse, 1957; Read & Leigh, 1967).

2) The response of ventilation to increasing PCO_2 becomes progressively more steep as the PO_2 is lowered, but the threshold (extrapolated PCO_2 at zero ventilation) remains the same. The raising of PO_2 conversely diminishes the response, but the effect reaches a limit at about 120 torr. There remains a response to CO_2 at all oxygen levels above this, presumably representing an entirely

central stimulation. It has however been reported that the response to CO_2 is depressed still further by increasing pressures of oxygen up to and above 1 atm (Lambertsen, 1963).

3) The response of ventilation to hypoxia can be expressed as a family of hyperbolic curves for different PCO_2 levels, derived from the responses described above.

4) The whole CO_2-ventilation response curve, and the whole "fan" of curves for different PO_2's can be shifted, i.e. a given PCO_2 can be associated with a higher or lower ventilation level, yet the sensitivity (slope of the response curve) be unaltered by a number of agents such as acidaemia-alkalaemia, sleep, exercise, drugs (Lloyd & Cunningham, 1963).

Information from timed breath-by-breath studies following transient or abrupt changes in inspired gas

Dejours (1957) advocated the use of a transient change in inspired gas concentration as a suitable method of studying responses in man to chemoreceptor stimulation. He pointed out that the initial disturbance of the status

quo, rather than a "steady state" response involving secondary as well as primary effects, was most useful in studying the response mechanisms. He has not been alone in using such methods, although the work of his group has been the most extensive. In a painstaking study on a single subject, by Hornbein, Roos & Griffo (1961), it was established that there was a ventilatory response to transient changes in oxygen tension, even of the order of only -10 torr.

Dejours studied three normal subjects, at rest and during exercise, at sea level and at high altitude, and showed that if there was a background of hypoxia, or even of normoxia (air-breathing) there was a definable depression of ventilation brought about by only one, or two, tidal breaths of oxygen; this occurred in the 3rd-4th breath after the beginning of the first intake of oxygen, or after a time interval of 10-12 sec (Dejours, Labrousse, Raynaud, Girard & Teillac, 1958). This they were able to label peripheral chemoreceptor response by comparing it with that which could be demonstrated in intact dogs, and could be shown to be abolished by chemodenervation (Bouverot, Flandrois & Grandpierre, 1961). Thus the presence of a hypoxic drive, even in eupnoea and

normoxia, was demonstrated in normal man. Conversely, one or two tidal breaths of nitrogen or of CO_2 caused a stimulation of breathing with a similar time course. Dejours himself pointed out that the extent of the depression, or of the stimulation, could not by this method be precisely related to a particular step change in the blood gases, but he did establish the time relations of an effect attributable to the chemoreceptors. Fitzgerald and co-workers took up this point and applied transient stimuli in the form of 'slugs' of altered blood, to the carotid region of dogs, imitating the changes produced by one or two breaths of altered inspired gas; they found ventilatory responses compatible with those of Dejours (Fitzgerald, Zajtchuk, Penman & Perkins, 1964). More recently Leitner et al (1965) have demonstrated a close relationship in timing and extent between ventilatory changes and chemoreceptor impulse traffic during this type of 2-breath test.

Studies by Lambertsen and his colleagues have also included examination of the time course of responses to changes in inspired gases, in awake dogs and in a few human subjects (Gelfand, Lambertsen & Davies, 1964; Lambertsen, Gelfand & Kemp, 1965). Breath-by-breath

recording of tidal volume and frequency, and of minute volume per breath, led them to define at least three components of the response to CO_2, with different delays from the first breath. Their value for delay time for the fastest component seems much shorter than that of Dejours, but understandably so when it is realised that their studies are all carried out at raised controlled CO_2 levels (Lambertsen & Wendel, 1960): the background ventilation was very much higher, so mixing of altered inspired gas would be more rapid, as also would transit time to the chemoreceptors.

Approach to the present experiments

On leaving CO_2 uncontrolled

Observations have been made on resting man. He is notoriously a difficult subject: his heart rate and his ventilation, in particular, are liable to vary with the slightest disturbance of his external or internal environment. Nevertheless it has been possible, with subjects used to familiar laboratory surroundings, to obtain repeatable steady base line levels of ventilation and heart rate and to demonstrate induced changes which are significant in spite of being small. Because earlier experience was encouraging in this respect, such experiments were continued, following a preference for observing effects on the whole organism without the introduction of artificial control of secondary changes, which might obscure more than it clarifies.

It has become such a standard procedure to study responses to hypoxia, or to hyperoxia, at controlled, elevated P_ACO_2, after the example of Lambertsen and his colleagues, or to add a small percentage of CO_2 to the background "air" in order to create a steady baseline ventilation level, as by Cunningham's group, that to study straightforwardly what happens when a man is exposed only

to altered oxygen tensions, has become almost reactionary. And indeed, such observations are more complex in the sense that the single primary effect and the secondary effects of the response to it have to be disentangled in interpreting results.

But the attempt to hold all stimulating factors but one constant, in the classical experimental style, rather breaks down for whole humans when a factor controlled is P_ACO_2: because such control does not hold steady the ultimate CO_2 stimulus to ventilation.

Suppose that two levels of P_AO_2 are being compared for their effects on ventilation at two fixed levels of P_ACO_2, both above normal: the higher PCO_2 will give a greater increase in CBF and therefore a relatively higher cerebral PO_2 than will the lower PCO_2. The lower PO_2 will enhance the vasodilatory effect of either PCO_2 more than will the higher PO_2; there will be therefore less difference between the two PO_2's in the cerebral tissue than between the two arterial or alveolar levels. These factors have been appreciated, and introduced into sophisticated mathematical models of the control system (Grodins, 1964; Read & Leigh, 1967) yet maintenance of

steady alveolar PCO_2 is still used on the assumption that it holds one controlling factor constant.

In the study of circulatory responses, there is evidence that the small secondary changes in PCO_2, which follow changes in inspired oxygen concentration in man, do not in themselves lead to any significant modification of the responses (Richardson, Kontos, Shapiro & Patterson, 1966). This was not the view of Black & Roddie (1958) but they did not accurately match blood gas levels in their comparisons. Neither, however, does it seem to fit in with the great sensitivity of cardiovascular variables to small rises in PCO_2, as reported for example, in dogs, by Noble, Trenchard & Guz (1967).

In either case, whether cardiovascular or ventilatory variables are studied, it seems appropriate in the assessment of the responses of whole man, to apply only one primary stimulus, and to observe its effects if possible before the secondary changes have had time to develop. This is the approach of Dejours.

On the combined observation of cardiovascular and ventilatory changes

All too often the two "systems" are studied separately. Yet to consider a change in ventilation without

considering concurrent changes in the circulation, is like considering the efficiency of travel to Paris in terms of London-Dover timetables only. Ventilatory and cardiovascular homeostasis have their crucial purpose in common-gas exchange between atmosphere and tissues. To what extent are the systems linked in their regulation? In 1939 Gesell predicted that they should be very closely linked; more recent research has tended rather to demonstrate their separateness. Causes and effects in the relationships between the controlling mechanisms of the two have received little attention: description of respiratory rhythms in heart rate and in blood pressure appear from time to time, but have conflicting explanations whereas close observation of the sequence of the several changes might settle the feasibility of certain theories.

Present experiments

Observations have been made concerning chemical influences on ventilation and heart rate in normal man at rest. Earlier work was concerned with ventilation only; later, heart rate was observed in addition. The responses studied were those for which no completely adequate information or explanation is available, or for which there is no confirmation, in man, of findings in animals. They are all related to the rôle of the arterial chemoreceptors; the basic approach has been to study the time-course of responses during changing states induced by sudden alterations in the composition of inspired gas. Throughout the period in which this work was done, instrumentation has been improved; the third problem in the thesis (section 5), that of the tachycardia in hypoxia, was dealt with, in most detail, both experimentally and analytically; the first two problems, that of the response to CO_2 in hyperoxia, and the response to hyperoxia itself, are presented more briefly: they represent the development of a line of thought, and an experimental approach, whilst also adding some information about normal responses.

It is inherent in the nature of the subjects, of the experimental situation, and of the problems considered, that ultimate answers were not likely to be found with the tools available. The value of the observations is rather, it is hoped, to demonstrate whether or not the time course of the responses in man would, or would not, fit certain hypotheses. If the primary effect of a certain stimulus is said to be x, but will be rapidly obscured by the result of its own secondary effect, y, then perhaps it may be possible to observe x before y happens. If on the other hand x is commonly held to lead to y, and one observes that y as often precedes x as follows it, then the proposition cannot be supported. If x and y happen concurrently one is no nearer the answer, unless there is a measurable time necessary for x to cause y. This approach seems to have been used surprisingly little in the validation of physiological hypotheses: "still" pictures before and after certain changes are studied, rather than the sequence of changes.

Summary of experiments

1) the effect of CO_2 in oxygen, compared to that in air, was studied by a double breath method similar

to that of Dejours in order to demonstrate by the timing of any response, whether or not there was a peripheral chemoreflex component. (Section 3).

2) the effect of 100% oxygen on ventilation, and on heart rate in a few studies, was observed breath-by-breath in the first minutes in order to define the relative timing of changes. (Section 4).

3) the effect of 10% oxygen on heart rate as well as on ventilation, was studied in order to discover which of the possible explanations of the mechanism of the production of tachycardia would fit in with the observed sequence and time delays. (Section 5).

GENERAL METHODS

Measurements of ventilation

a. Pneumotachography

Principles

Problems

Present method

b. End-tidal PCO_2

Sampling

Analyser

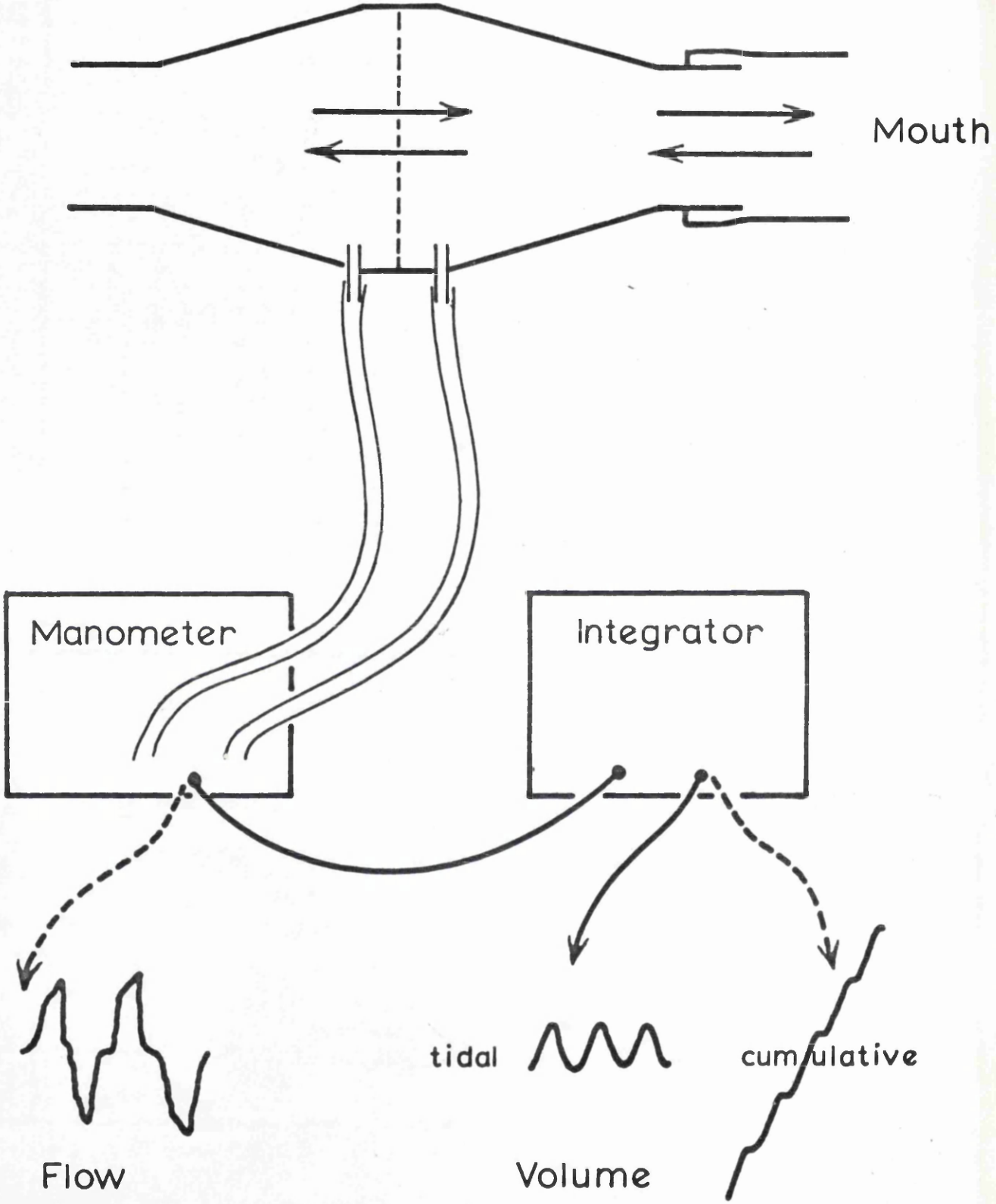
Ultra-violet recorder

Supply of inspired gases

Oxygen analyser

Fig 2:1

Principle of pneumotachograph



GENERAL METHODS

The essential components of the apparatus used for all experiments will be described in this section. Particular modifications or additions and details of procedure will be dealt with in their appropriate contexts later.

1. Measurements of ventilation

a. Pneumotachography

Principle: The pressure drop across a low-resistance screen (flow head) placed in the stream of respired air is proportional to the flow, if the screen is of suitable material and size. The flow can be electronically integrated with respect to time, such that the integrator output is proportional to the volume which has passed since the point of zero flow. When the flow head is placed in the to-and-fro airstream at the mouth, the integrator may give an in-and-out tidal output; alternatively a cumulative device will summate the volumes passing in one direction only, over a selected period (Fig. 2:1).

Fig 2:2

Trace obtained from integrator during one-way flow through flow head by suction from pump with constant stroke volume.

AIR O_2 : Model constant V_I





At the arrow the "inspired " gas was changed from air to oxygen.

Problems and potential errors

i) Viscosity of gas: A higher viscosity will give a greater pressure drop across the flow head, and so a greater apparent volume. If the pneumotachograph is calibrated with air, a correction therefore has to be made of volume obtained, e.g. with pure oxygen (Fig. 2:2). The differences between inspired and expired air are not significant in this respect.

Temperature causes a significant change in viscosity, such that the difference between inspired air at room temperature and expired air would strictly require correction: this temperature difference would vary with changes in ventilatory volume, and would alter during rebreathing from a closed circuit. The viscosity change due to differences in humidity is negligible (Greer, 1966).

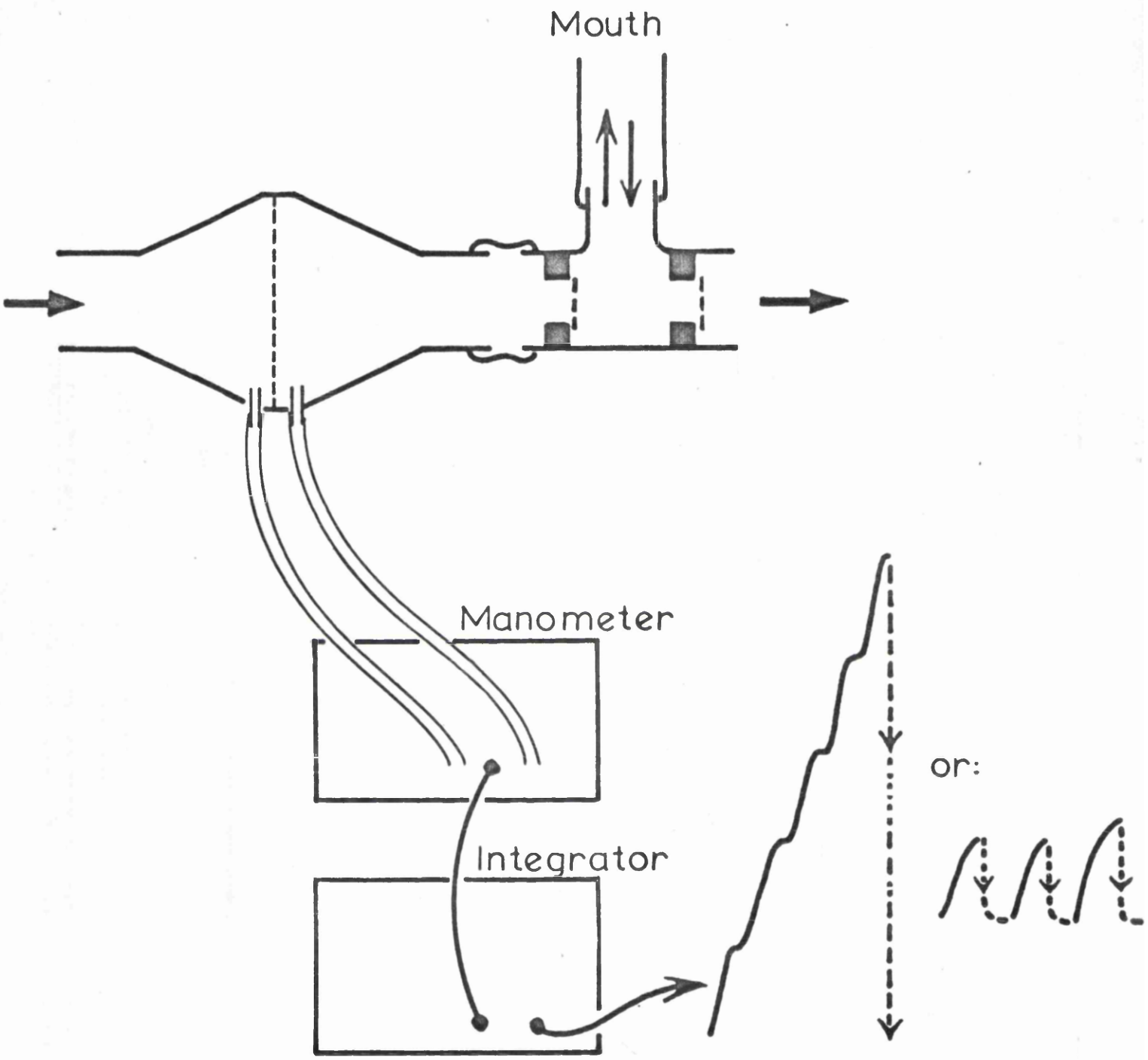
ii) Dead space: if a pneumotachograph is used in the to-and-fro position, considerable dead space is added.

iii) Condensation: expired air passing through a cool flow head inevitably leads to this. Also a flow head placed lower than the mouth collects saliva. A heating coil is provided in some flow heads.

iv) Accuracy and Calibration: The particular flow head used for given experiments must be one whose range of linearity spans the range of air flow rates likely to be encountered. Flow range during resting breathing is up to 40-50 L/min. An increase of ventilation rate to 40 L/min, which might occur for example in a CO_2 response test, would incur flow rates of up to 150 L/min. Exercise studies require larger diameters of screen than resting studies. There may therefore be difficulties in the transitions of ventilation from resting to stimulated conditions. Calibration for flow is relatively straightforward: different constant flow rates are put through the flow head using rotameters to test the linearity of the output. Linearity of integrated volume is dependent on the electronic arrangements of the particular integrator, and should be reliable if flow:pressure difference is linear. Triggering for the start of one-way integration has to depend on a pressure difference near to zero: this must be finely adjusted in order not to "lose" some of the flow before integration starts, or after it ends, especially

Fig 2:3

Inspiratory pneumotachograph



in relatively slow breathing. All these considerations make it advisable to assess the accuracy of the whole apparatus for volume measurement during different breathing patterns, actual or simulated.

Present approach to these problems

Problems i - iii are simply answered by placing the flow head in the path of inspired air (Fig. 2:3): temperature need not vary; changes in composition of inspired gas are known and the necessary correction is calculable; the dead space is that of a valve box only; condensation from expired gas is excluded.

With the flow head so placed it is necessary to have an integrator which will automatically return to baseline either after a selected cumulative deflection, a selected time, or after each breath.

The present method

Flow head: In earlier experiments a Fleisch Type 2 was used, and more recently a new type of light-weight flow head consisting of a single wire mesh screen mounted in a plastic housing (Mercury Electronics).

Electromanometer: The instrument used to measure the pressure difference across the flow head was the

photo-electric pressure transducer of Mercury Electronics (Greer, 1958).

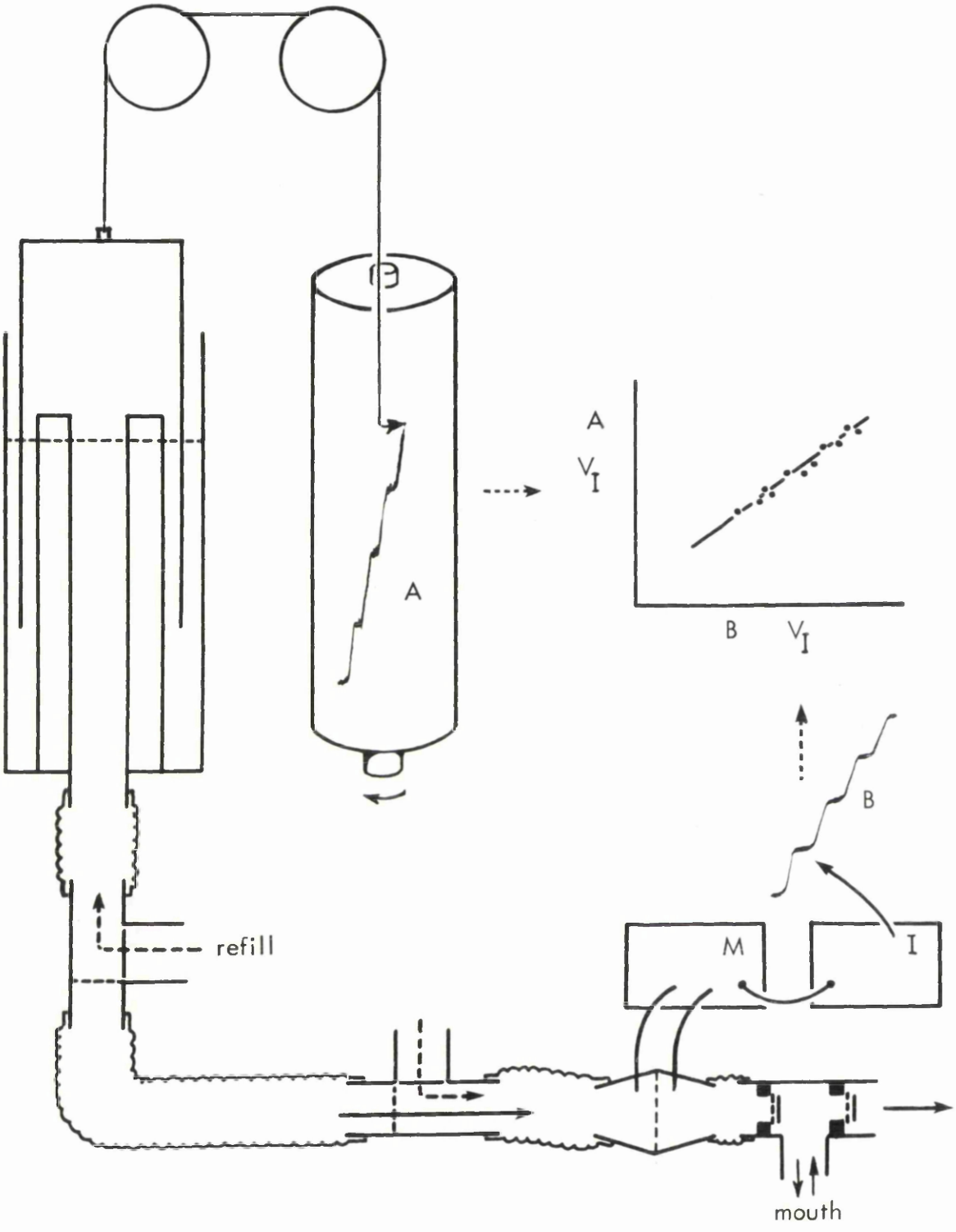
The manometer output was linear for constant flow rates in the range 20-160 L/min.

Integrator: Initially a tidal integrator was used (Greer, 1966) with the flow head at the mouth. Because of the disadvantages discussed, modifications were suggested. During the time that these studies were in progress, a new type of manometer-integrator was under development and repeated modification by Dr. Greer. A cumulative output was used in some earlier experiments, but later the alternative facility was used of an output which returned to base-line after each inspiration. Ultimately a breath-by-breath ventilation-rate computer was added, and will be described in section 5 in relation to the experiments for which it was used.

Calibration of integrated volume output

Linearity of the recorded volumes was checked in two ways: in an earlier arrangement which applied to the experiments in section 3 and some of section 4, a subject was connected to a circuit containing both spirometer and pneumotachograph; volumes of individual breaths obtained by each method were plotted against each

Fig 2:4



Breathing circuit for correlating inspiratory volumes measured by spirometer and by pneumotachograph.

other. Deliberately wide variations in tidal volume, and in velocity of inspiration were used, and no significant non-linearity was found (Fig. 2:4).

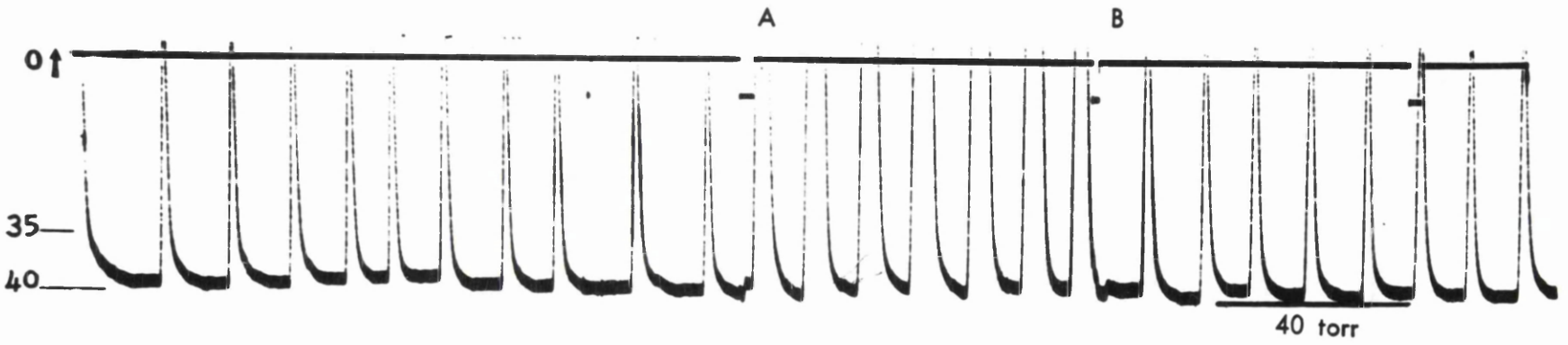
Later a mechanically driven spirometer, set to different frequencies and tidal volumes, was set up to check linearity when any new flow head of manometer-integrator arrangement was used.

In the experiments to be described, there was not inherently any advantage in knowing the absolute values for ventilation, since relative changes provided the essential information. Arbitrary units were therefore mostly used. In some experimental results absolute values are quoted: these were derived from concurrent measurements of expired volume by a dry gas meter. Otherwise the response of any subject on any occasion was related to his baseline ventilation on that occasion. Levels of ventilation were only compared between subjects if the experiments had been carried out in a group using similar instrumentation, and with similar setting of controls. The normality of ventilation in any instance was assessed from the end-tidal PCO_2 level.

b. End-tidal PCO_2: This was continuously followed by sampling and analysing gas either from immediately beyond the

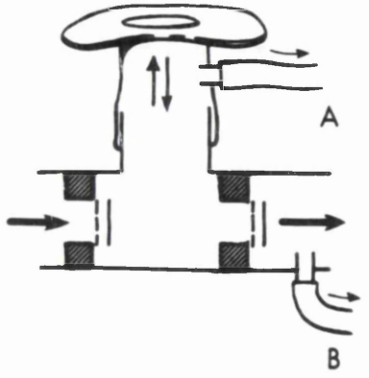
Fig 2:5

Trace of continuous CO_2 analysis; comparison of two different sampling sites



Increasing CO_2 downwards

expiratory ↑ mouthpiece ↑ expiratory



A : Mouthpiece
B : Expiratory

sampling points for CO_2

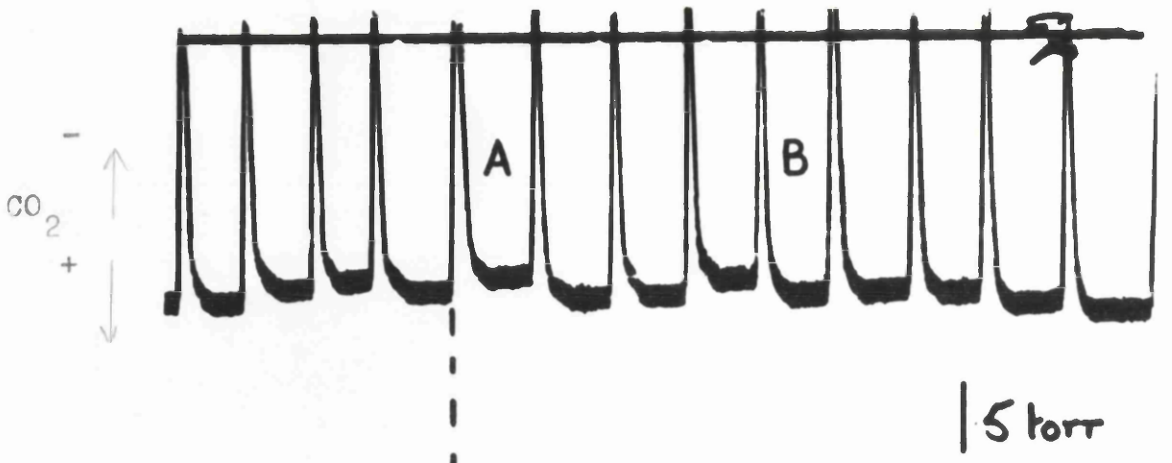
expiratory valve, or from the mouthpiece.

When the sampling site is at the mouth, in both inspired and expired air streams, a plateau level towards the end of expiration can be taken to represent the alveolar CO_2 concentration: if the breath is too small, or the time too short, it is clear from the trace that the peak value cannot be taken to be the alveolar value, because it will fail to level off.

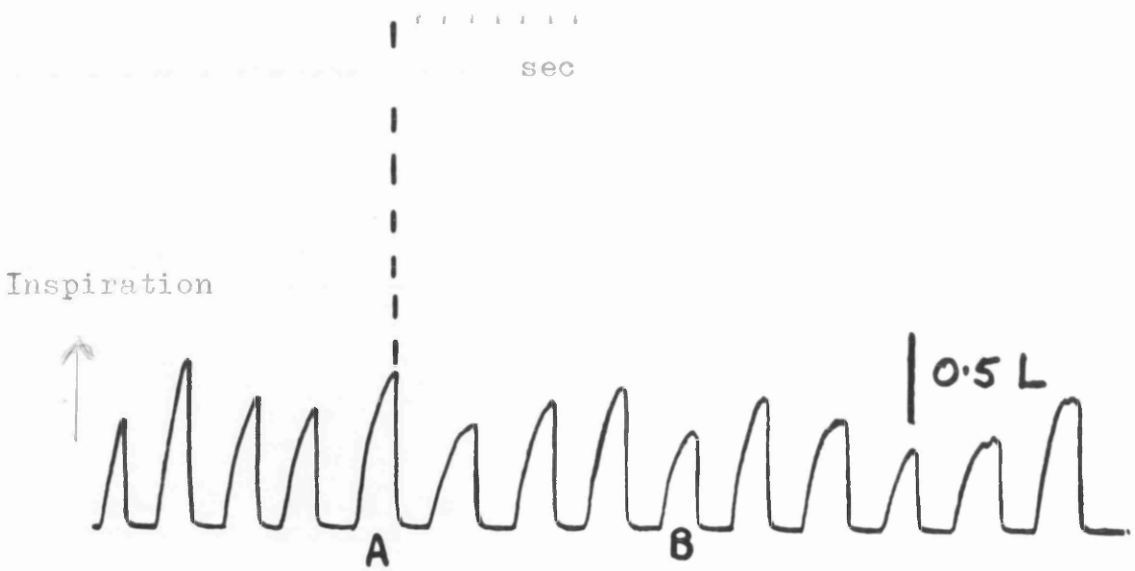
The expiratory site is useful if sampling is carried out in conjunction with collection of expired gas, because the sample can be returned downstream; it has the disadvantage that a larger minimal tidal volume is necessary for sufficient alveolar gas to reach the sampling point, and that continued intake of end-tidal gas during the succeeding inspiration gives a false plateau, which does not necessarily represent the alveolar value. In order to be sure that alveolar plateau has been reached, it is necessary to identify the point at which inspiration began, from the concurrent tidal trace. Fig. 2:5 shows a comparison of the traces obtained by the two methods in continuity: a trained resting subject breathed steadily whilst a switch was made from the one sampling site to the other.

The analyser used was the URAS infra-red (Godart C.P.I. Ltd: Hartmann & Braun). CO_2 can be read to an accuracy of 0.5%. The response lags 1 - 2 sec (Fig. 2:6) behind a change of CO_2 concentration at the sampling site, according to the length and diameter of the sampling tube and the flow rate; the response of the instrument itself, when the front of altered gas enters the analyser inlet, is 0.6 - 0.8 sec again depending on the flow rate. It is

Fig 2:6



Tidal volume and $P_A CO_2$



Upper trace : CO_2 sampled from beyond expiratory valve.

Lower trace: Inspiratory tidal volume.

A...A, B...B : corresponding inspired volume and expiratory analysis

therefore rapid enough accurately to follow breath-by-breath changes. Fig. 2:6 shows an example where expiratory sampling was used. The fall to zero represents dead space air passing the expiratory valve. The trace also shows the close inverse relationship between the depth of inspiration and the end-tidal level of the succeeding expiration.

Calibration of the analyser was carried out before each day's experiments, using two gas mixtures containing known percentages of CO_2, analysed on the Lloyd-Haldane apparatus. In earlier studies, calibration was also checked at frequent intervals during experimental runs. It was found, however, that our particular instrument, continuously switched on, remained stable, and did not require recalibration during a session.

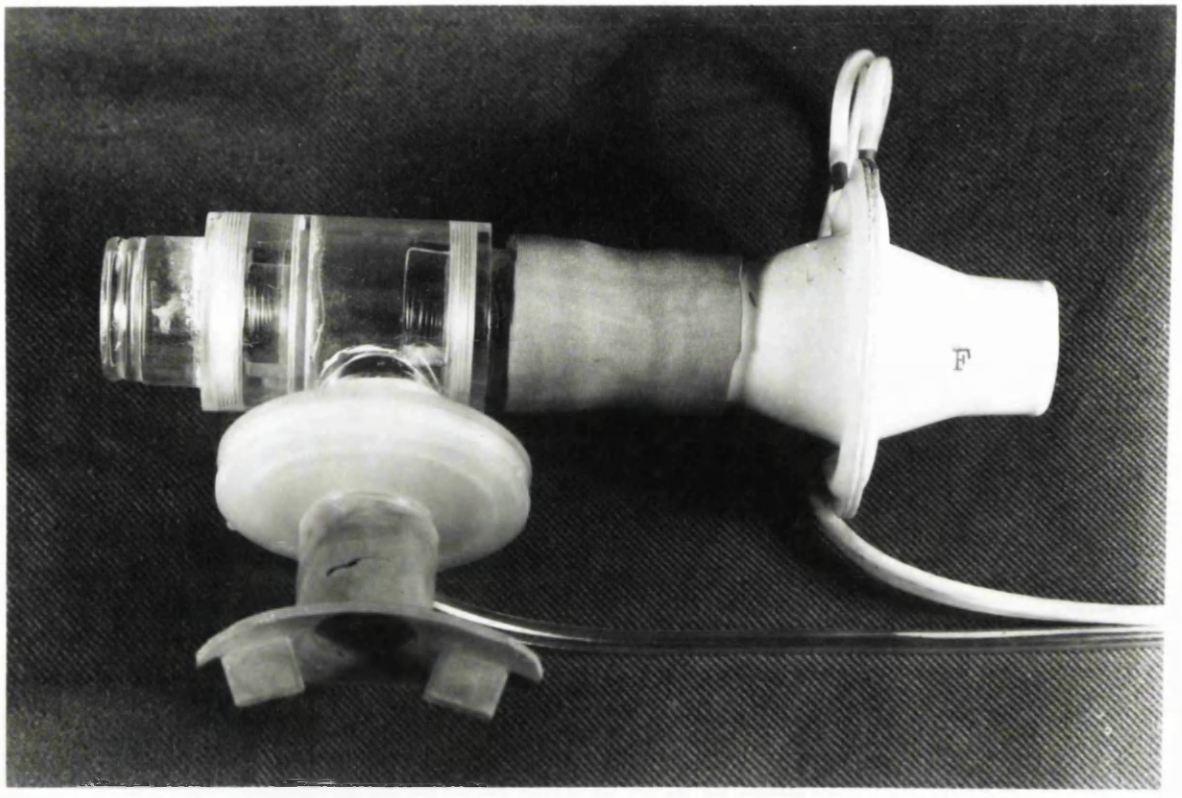
Sample flow rate: Usually the intake to the analyser was kept at 40L/hr. Analysis was always carried out at the same sample flow rate as calibration.

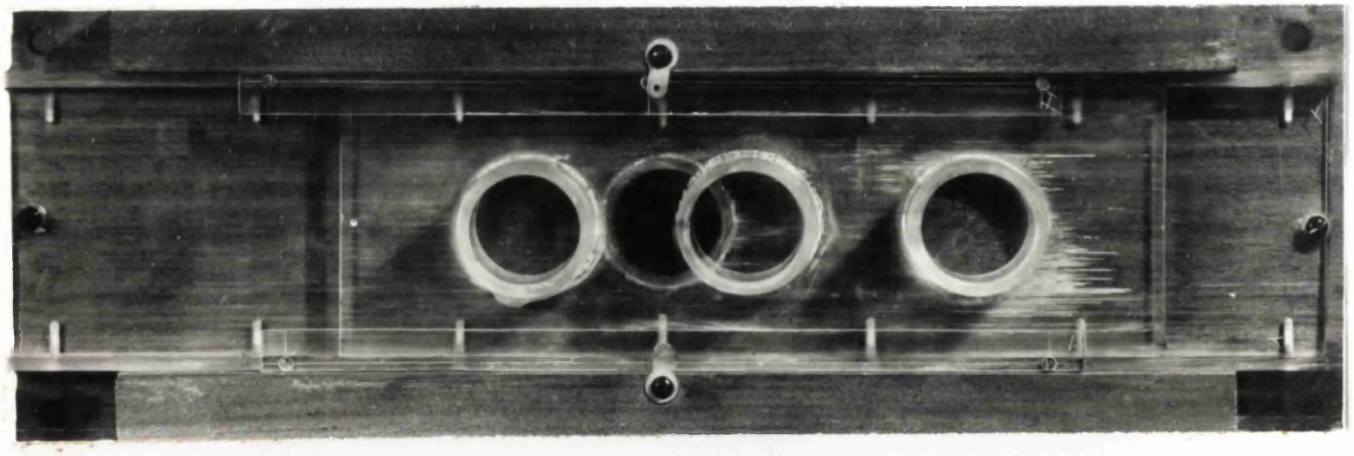
Carrier gas: Calibration was with gases approximating in oxygen content to expired air. The effect of oxygen as the carrier gas was tested by analysing CO_2 mixtures in oxygen volumetrically, and comparing the reading of the

Fig 2:7

Above : Flow head (F)
Perspex valve box
Condenser-humidifier
Mouthpiece

Below : Sliding valve





infra-red analyser. Repeated estimations using different percentage CO_2 showed a difference rather smaller than that quoted by the makers. According to our own observations the correction required amounted to -0.01%(< 0.1 torr) for a reading of 5% CO_2 in oxygen when the instrument had been calibrated with 16% O_2.

Ultra-violet recorder

Initially, two galvanometers of a multichannel U-V Recorder (S.E. Laboratories) were used for ventilation and CO_2 recording. Later a third galvanometer was used for instantaneous heart rate, and a fourth for breath-by-breath ventilation rate (p. 5/25)

A remote control event marker was added so that a single operator could change inspired gas and mark the chart record. The response time of this recorder is faster than the minimum required for recording from the pneumotachograph, CO_2 analyser or ratemeter.

Supply of inspired gases

Gases were supplied from PVC Douglas bags (Plysu Ltd.) through flexible hosing, attached to one of the three inlets of a sliding valve, made for this purpose in our own workshop (Fig. 2:7). This enabled unobtrusive

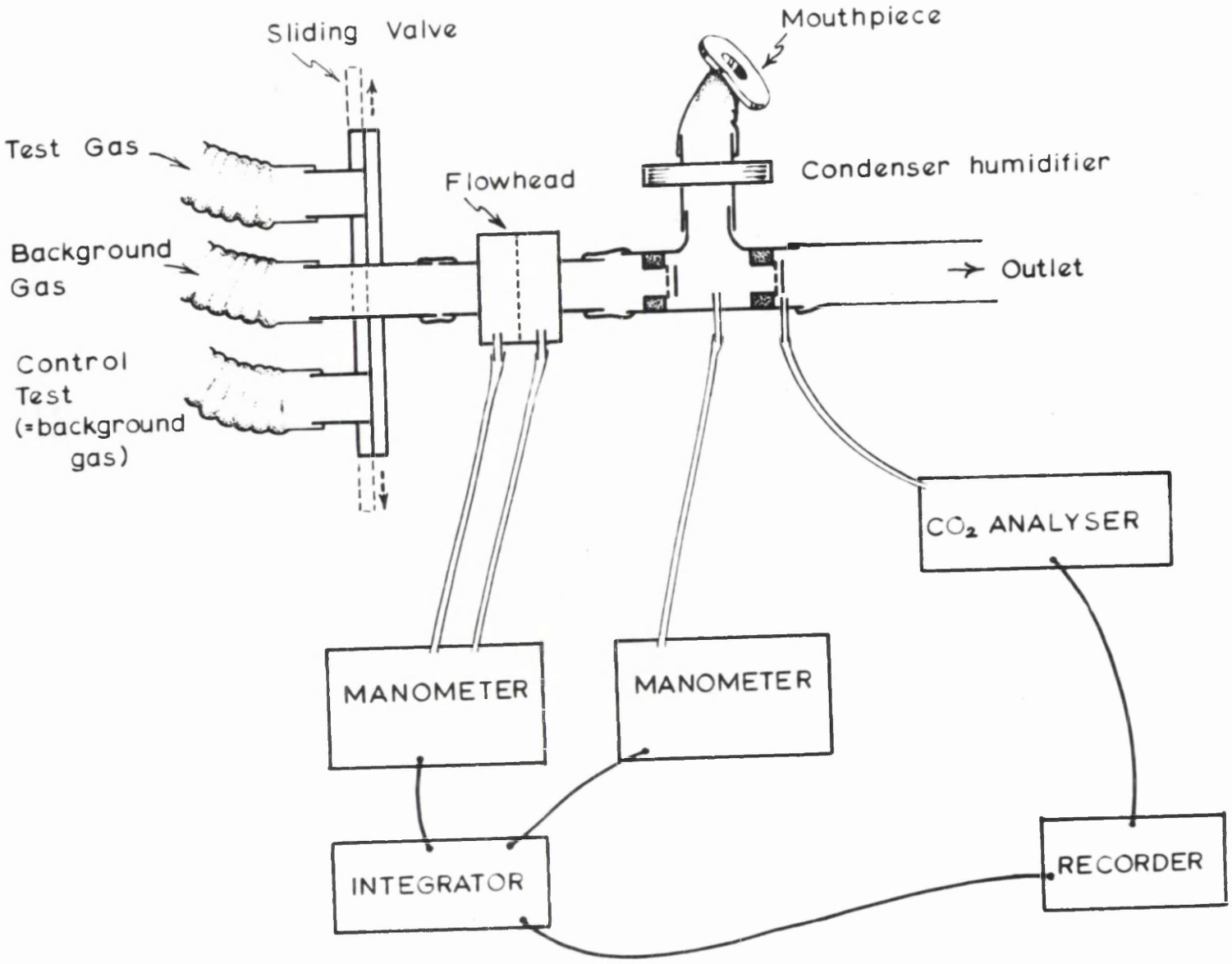


Fig 2:8

Arrangement of apparatus used in earlier experiments.

A later version is shown in Fig 5:1.

changes to be made, by sliding the appropriate inlet into position opposite the single outlet.

Bags were filled well in advance of experiments to allow equilibration with room temperature.

The subject was connected to a mouthpiece, perspex valve box, and thence on the inspiratory side to the flow head and the slider. A condenser-humidifier (Garthur) was interposed between mouthpiece and valve box to minimise dryness in many of the longer experiments (Fig. 2:8).

Dead space

Most experiments involved the study of a step-change in inspired gas composition. The apparatus dead space between the slider and the mouthpiece was therefore kept as small as possible. This was 95-98 ml, by repeated measurement. The hosing from any Douglas bag containing a mixture other than air was always flushed through with the gas before it was attached to the slider.

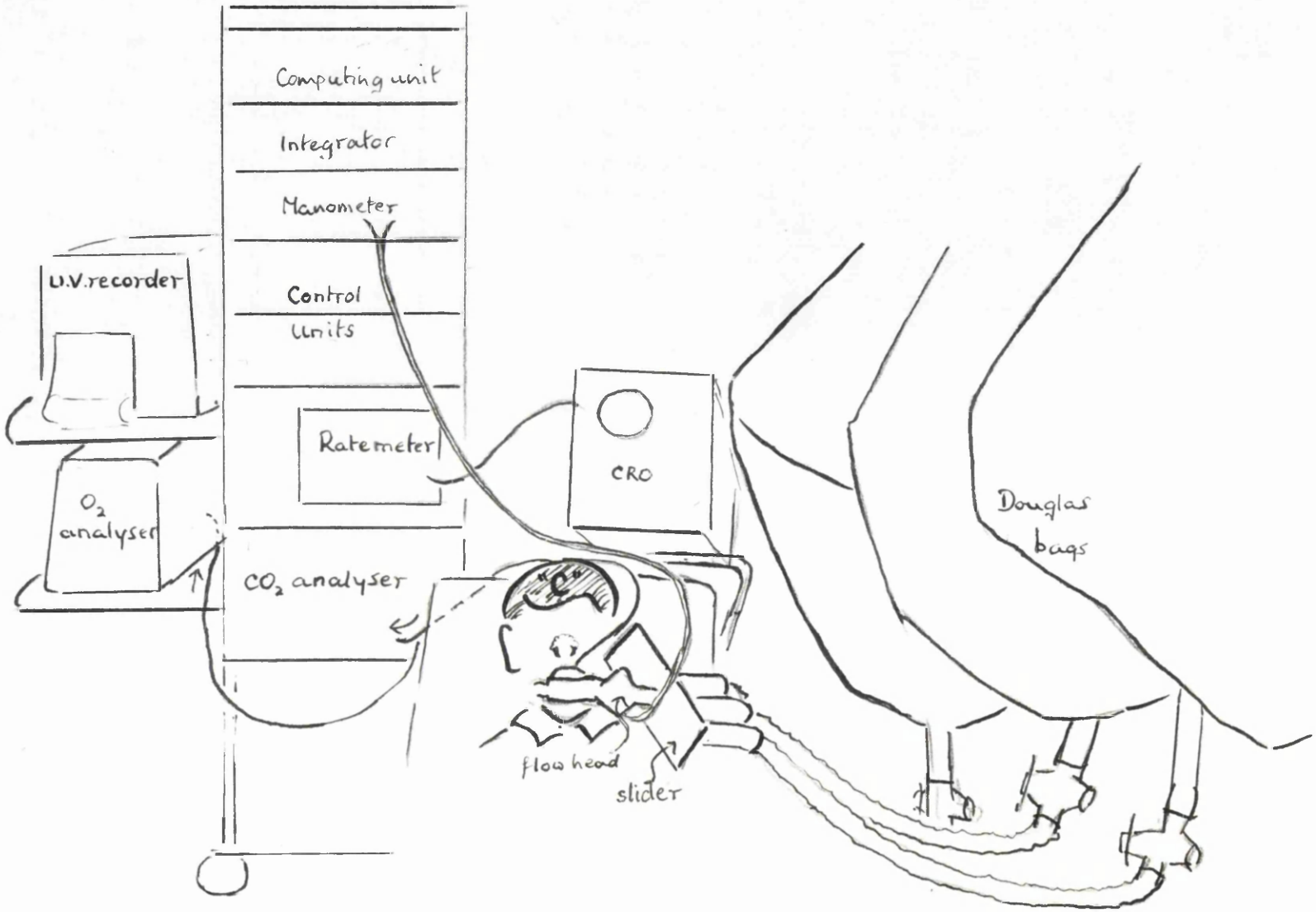
Resistance

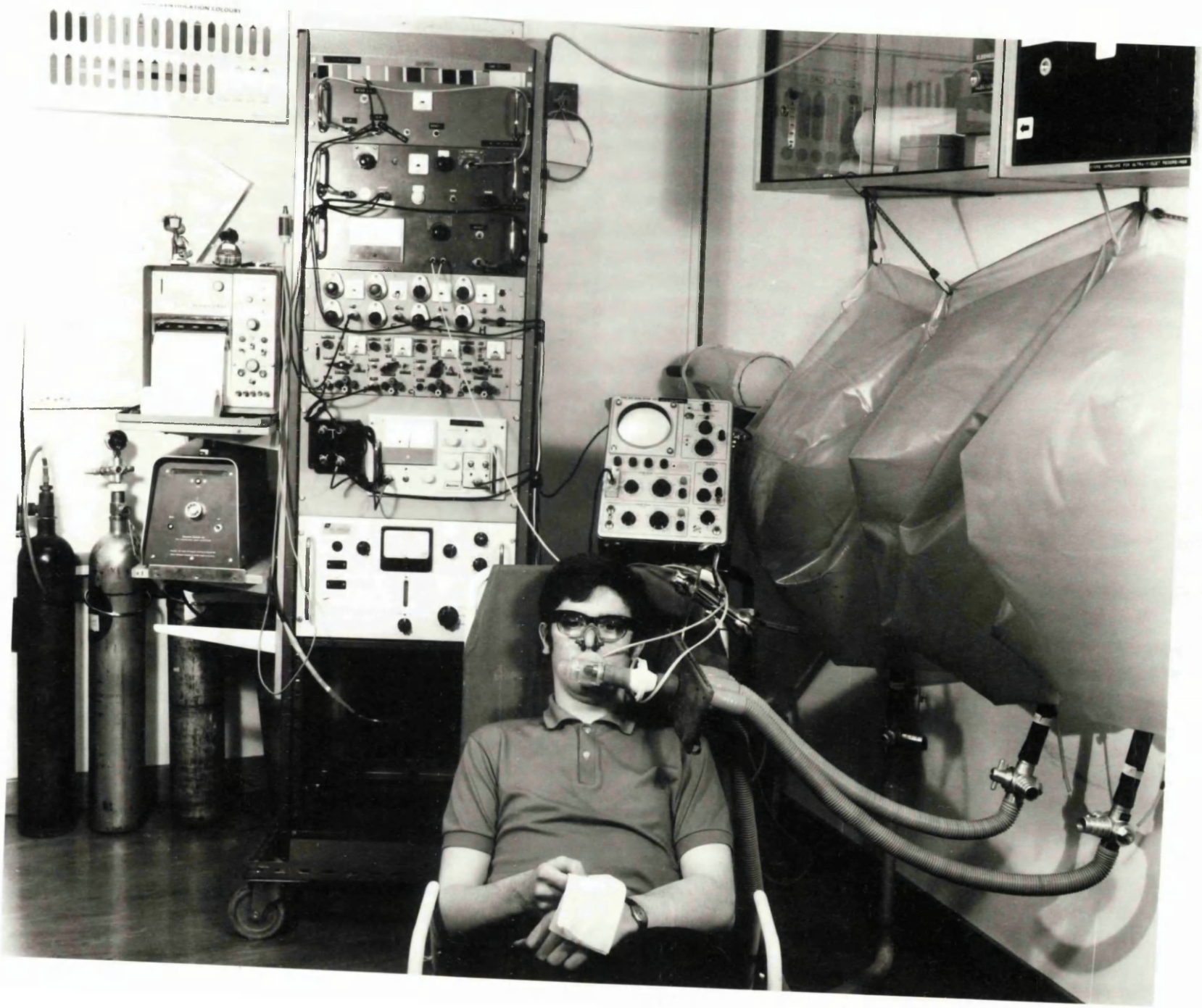
To assess this, pressures were recorded at the mouth during normal breathing through the assembled apparatus. The swing of pressure was approximately -0.5 to +1 cm H_2O.

Oxygen analyser

No facility was available for continuous oxygen analysis. For various purposes during different experiments it was necessary to analyse samples for oxygen. The Servomex paramagnetic analyser was used (Type DCL Mark II). The zero reading was checked on every occasion with pure nitrogen. The span was adjusted by calibrating with gas mixtures close to the composition of expired air, or of other mixtures to be analysed.

The present form of the combined apparatus is illustrated overleaf.


Computing unit
Integrator
Manometer
U.V. recorder
Control
units
Ratemeter
CRO
Douglas
bags
O_2
analyser
CO_2 analyser
flow head
slider



HYPEROXIA AND THE CHEMORECEPTOR RESPONSE TO CO_2

Timing and extent of the ventilatory response to transient increase in inspired CO_2 concentration against background of air and of pure oxygen respectively

Introduction

First series of experiments

- Method, double-breath CO_2 tests
- Subjects
- Procedure
- Results
- Discussion

Second series

- Methods - improvements on first series
- Results

Discussion

Conclusions

HYPEROXIA AND THE CHEMORECEPTOR RESPONSE TO CO_2

Timing and extent of the ventilatory response to transient increase in inspired CO_2 concentration against backgrounds of air and of pure oxygen respectively.

Introduction

Joels & Neil (1961) demonstrated interaction between hypoxia and hypercapnia at the arterial chemoreceptors, and they have stated that they are "adamant that the response of the chemoreceptors to changes in blood PCO_2 is almost negligible" if the animal is breathing high oxygen concentrations (Neil & Joels, 1963). This seems to be established for experimental animals. Bouverot, Flandrois, Puccinelli and Dejours (1963) reported that in dogs high oxygen eliminates the immediate reflex effects of step changes in PCO_2 at the chemoreceptors; more recently the same group, studying afferent chemoreceptor activity as well as ventilatory response, demonstrated that the pattern of response in hyperoxia was similar to that after chemodenervation (Leitner, Pagès, Puccinelli & Dejours, 1965b).

In any study including the effects of hyperoxia

in man, a knowledge of reflex mechanisms which are or are not active in this state is relevant. It was therefore proposed to investigate directly in man the proposition that there is no arterial chemoreflex response to CO_2 in hyperoxia.

Reports of CO_2 response in man

Dejours and his colleagues suggested that response to CO_2 should be studied by a method similar to their "O_2-test" or "N_2-test" (p.1/14), and gave as an example the mean results of 7 experiments: the ventilation could be seen to increase in the 2nd-6th breaths (about 8-30 sec) following a single tidal inspiration of 7% CO_2 in air (Dejours, Labrousse, Raynaud & Flandrois, 1958).

At the time when the present experiments were started, there was no comparable investigation reported comparing CO_2 in oxygen with CO_2 in air. Meanwhile Cunningham's group reported experiments using a different approach, to find an answer to the same question: subjects were studied in a steady state of high CO_2 and ventilation, with high or low oxygen. The time-course of ventilatory decrease after transient withdrawal of CO_2 from the

inspired gas was studied: the results indicated a rapid component when there was a hypoxic background, but not when there was hyperoxia (Cunningham, Lloyd, Miller & Young, 1965). Gelfand et al (1964) similarly used a raised controlled PCO_2 and studied off-transients. Several studies were done in dogs, and the time course of ventilatory response resolved into three exponential functions, of which the most rapid could be abolished by hyperoxia; their comparable studies in man had apparently only extended to one subject.

FIRST SERIES OF EXPERIMENTS

Methods

The effect on normal human subjects of two tidal breaths of 7% CO_2 in air, or in oxygen, was observed.

The first series of experiments was carried out recording ventilation only, before the acquisition of a rapid CO_2 analyser.

In order to establish the possibility of defining an arterial chemoreceptor response, many trial runs were carried out using single, double and triple-breath nitrogen tests. Many showed no response, retrospectively because of hyperventilation and low CO_2, so that the

hypoxia-hypocapnia combination was not reaching threshold level for stimulation. Selecting only those without obvious hyperventilation, it was possible to define a ventilatory response in the 4th-5th breath. A similar arrangement of apparatus was therefore used for CO_2 tests, in the hope of defining a response with a similar time course. The arrangement was as follows:

Ventilation was recorded continuously by inspiratory pneumotachography, as described. A Fleisch Type 2 flow head was used, and the integrator output used so as to give a cumulative, "staircase" trace on the UV recorder. At the limit of the paper the output was zero-ed by hand. There was a slight consistent non-linearity of the chart record in relation to the integrator output, such that a smaller apparent inspired volume was recorded at the top of the paper. This was allowed for by always using the 2nd-3rd breaths of a "staircase" for the test or control gas, so that any consistent artificial difference in the few breaths after the test would have been revealed equally in control tests.

Inspired gases were supplied from plastic Douglas bags, and changed by the sliding valve. For

any one run, three bags were filled with the background gas (air or oxygen) and one with the test gas (6-7% CO_2 with 21% oxygen in nitrogen, or with oxygen to 100%). The second bag of background gas was used for control "test", and the third for replenishing the background supply.

Subjects were 9 students, male, aged 19-21. Each subject reported at no particular time of day, but not within two hours of a heavy meal. He rested for 10-15 minutes, and during the latter 5 was accustomed to breathing through the mouthpiece, inhaling room air, seated, with most of the apparatus out of sight behind him.

Procedure

An air background and an oxygen background experiment were carried out on each subject on the same occasion. Subjects were randomly assigned to start with either the one or the other. This background was maintained for about 40 minutes, during which 16-20 double breath tests were applied: half by switching to the CO_2 mixture and allowing two tidal inspirations, half by switching to the control gas. The order of

control and CO_2 tests was determined randomly. At least 15 minutes rest was allowed between the two experiments. The positions on the slider of test gas and control gas were reversed about halfway through each experiment. At the end of each sequence of tests, the CO_2 mixture was switched on and continued for 2-3 minutes, before returning to background gas.

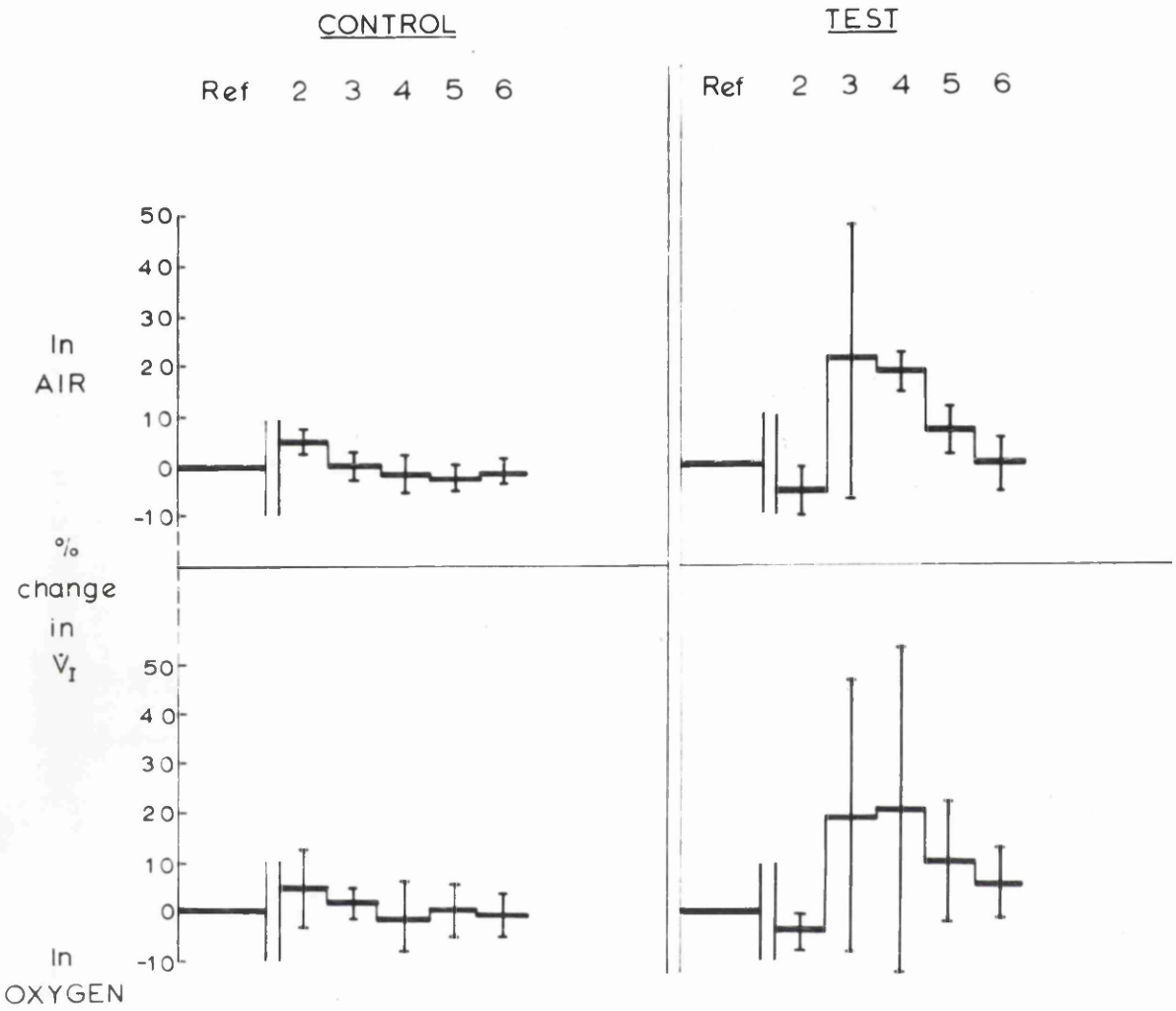
Results

Reference value for inspired volume (V_T) and for the time interval between breaths (t) was obtained for the 10 breaths preceding any test, giving a reference $\dot{V}_I$ value in arbitrary units ($\bar{V}_T \div \bar{t}$).

Values for 2nd to 6th breaths, after the change to CO_2 test or control test, were found so that "breath-by-breath minute volume" was obtained for each breath. t remained steady in many instances so that a value for V_T only could be used.

For each subject the mean ref. value for all tests in a run was found and the mean of all 2nd breaths, 3rd breaths etc. was expressed as a % of this, for all control tests, and for all CO_2 tests respectively. The results were expressed graphically as in Fig.3.2 overleaf which

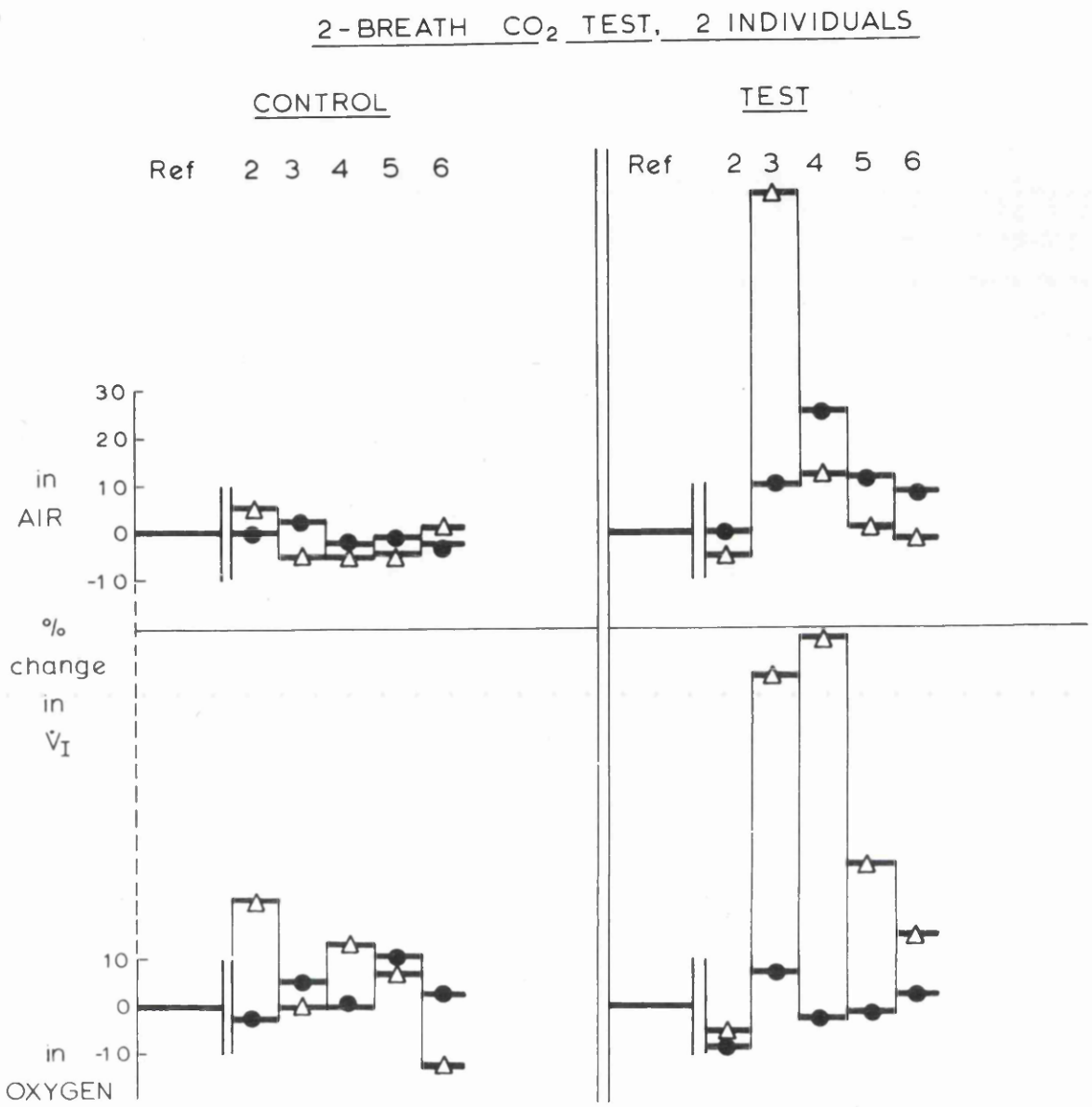
Fig 3:1



RESULTS from 7-10 CO_2 TESTS and CONTROL TESTS on EACH of 6 SUBJECTS

Numbers refer to 2nd...6th. breath of the test (see Fig 3:3,4).
Each value represents mean V_T/t for that numbered breath in all tests, expressed as % change from reference value (mean for $\frac{1}{2}$-min before test)
Bars show $\pm$ 1 S.D.

Fig 3:2



MEAN RESULTS of 7-10 TESTS on EACH SUBJECT

shows separate results for two dissimilar subjects. In those subjects (there were 3) in which no response was demonstrated on air, it was taken that they were ventilating excessively, and they were excluded.

Pooled results. All results for oxygen, and all results for air backgrounds respectively, were pooled.(Fig.3:1).

Air background

It can be seen that there was a clear difference between control and test data for these 6 subjects. The timing is similar to that quoted by Dejours et al. It can reasonably be taken that an arterial chemo-receptor effect is evident in the relatively constant response in the 4th breath.

Oxygen background

In some subjects there was a clear difference between the control and CO_2 tests (Fig.3:2), and the apparent response to CO_2 appeared to be in a chemoreceptor time pattern. Pooled results show the very considerable variability (Fig.3:1) between subjects, but suggest a similar pattern to the changes in air.

Continuous CO_2 breathing - see results of second series, p. 3/12.

Discussion

These experiments had the obvious disadvantage that the actual P_ACO_2 was unknown. It was therefore possible neither to know the background state of alveolar ventilation (correction of arbitrary units to L/min suggested high values for some subjects) nor to know the strength of the given stimulus in terms of rise of alveolar PCO_2. Nevertheless the results were suggestive enough of a positive response to CO_2 in high oxygen to justify a further series of experiments when a CO_2 analyser became available. These will now be reported.

SECOND SERIES

Methods

Ventilation was again recorded by inspiratory pneumotachograph, but a successive tidal output was used: by the time these experiments were done, the automatic breath-by-breath zero device had been incorporated, so that the record returned to baseline at the end of inspiratory flow (Fig. 2:3). Continuous infra-red analysis for end-tidal CO_2 (p. 2/7) was used.

Both were recorded continuously on two channels of the UV recorder.

Inspired gases were dispensed as in the first series, and the order of tests and controls was similarly randomised. However, as the difference between control and CO_2 tests had been established in the earlier series, fewer controls were included - 1 control to every 2 CO_2 tests.

Subjects were different from those of the first series: 5 young men (technical staff) were studied. Background oxygen was given for at least 4 min before starting tests. The procedure was similar, otherwise, to that of the first series, except that in addition a measurement of mixed venous PCO_2 by a rebreathing method, was added at the end of each experiment: these will be reported later (p. 4/13).

Results

Examples of the records obtained are shown in Fig.3:3,4; there was usually a response in normoxia.

Ventilation: In view of the experience of the first series, the results were processed in the following manner:

Fig 3:3

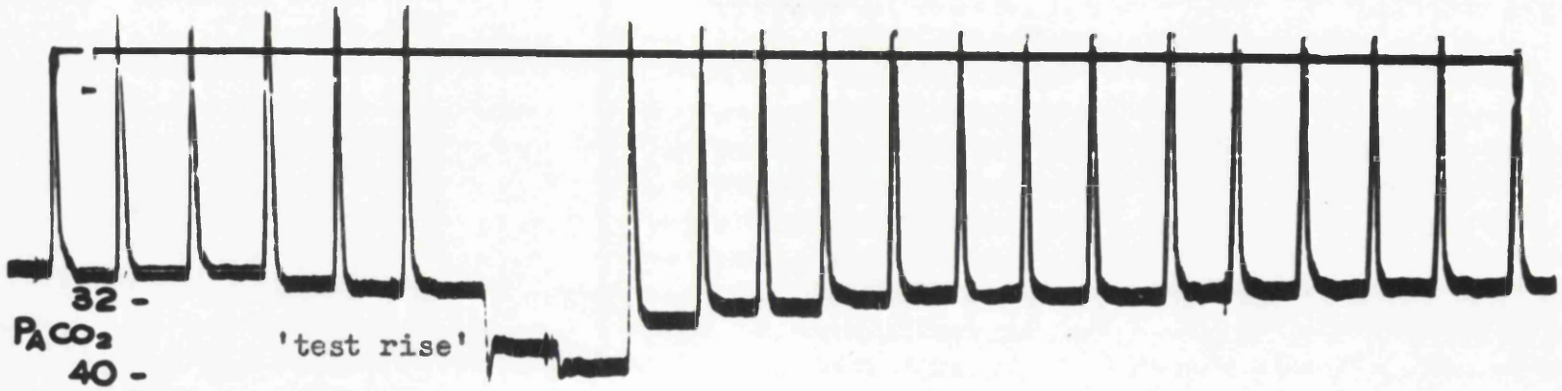
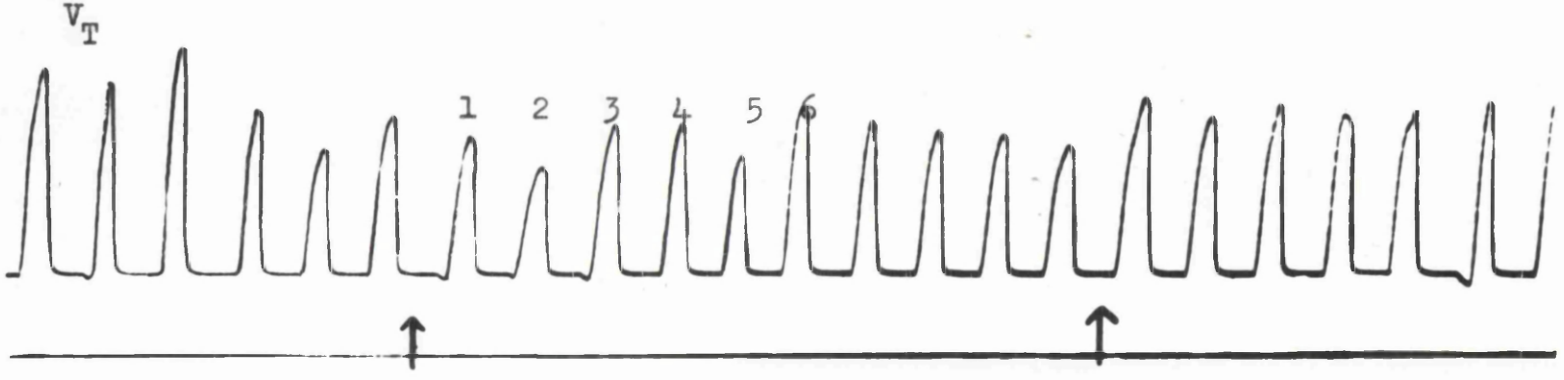


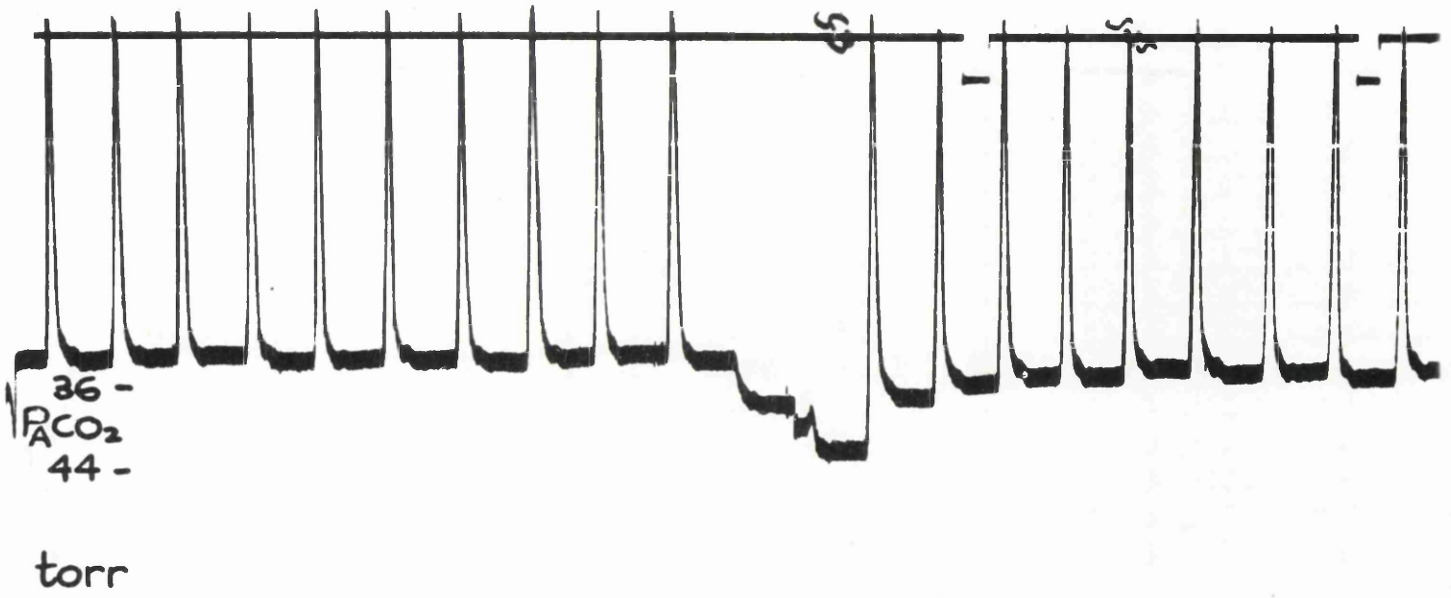
Fig 3:3

2-breath CO_2 test in O_2

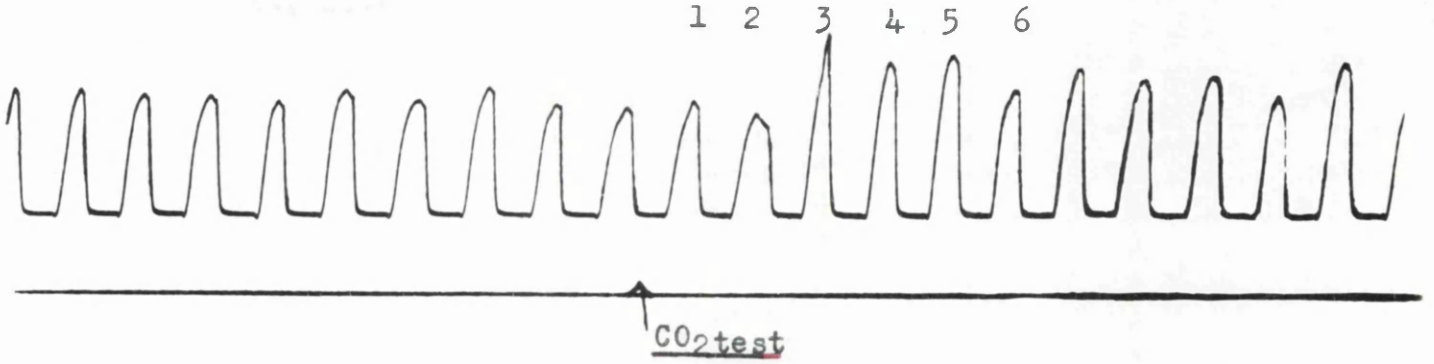


Subject N : background inspired gas, 100% oxygen. At 'CO_2 test', two breaths of 7%CO_2 in O_2. At 'control test', two breaths of O_2 from separate bag.

Fig 3:4



2-breath CO_2 test in AIR

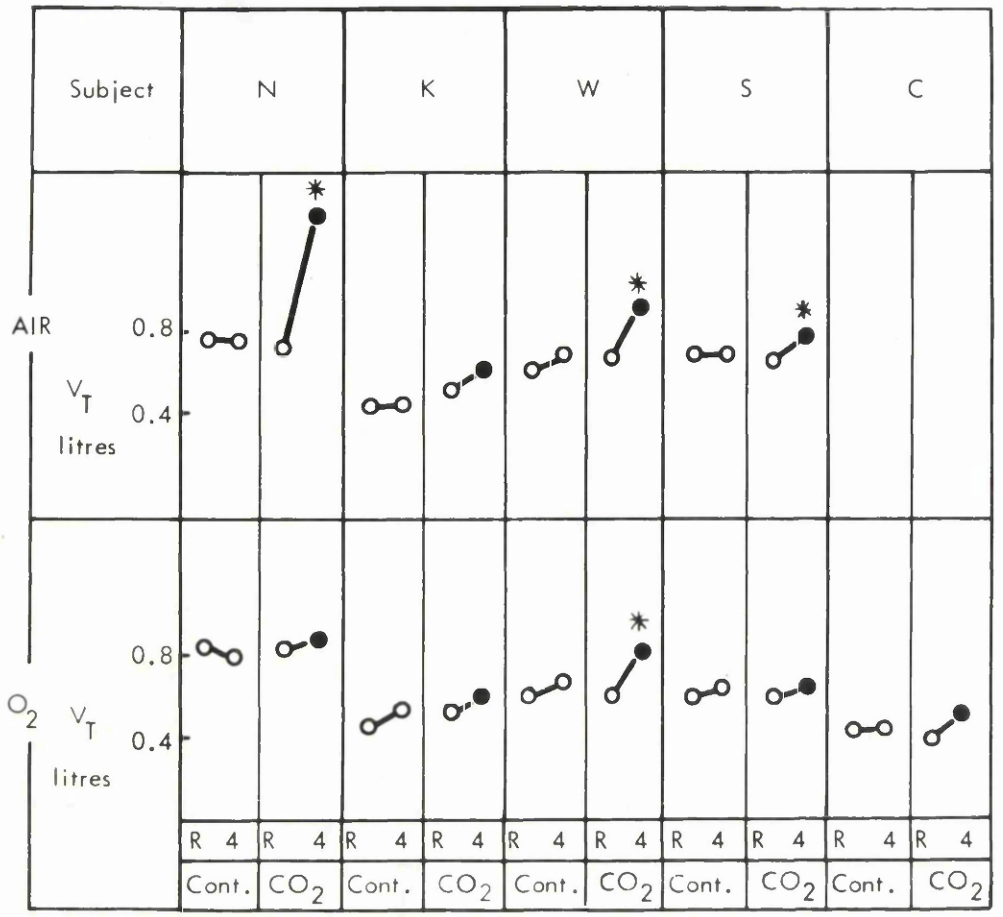


Subject N : same occasion as above. Background inspired gas : air. At 'CO2 test', $\frac{2}{3}$ two breaths of 7%CO_2 in a

Fig 3:4

DOUBLE BREATH TESTS

Increase of Tidal Volume in the 4th Breath



R - Reference value: V_T before test

4 - 4th breath after double breath test

Cont. - Control - double breath test with background gas

CO_2 - Double breath CO_2 test

* - Significantly higher than control

Fig 3:5

Double breath CO_2 and control tests, in air (4 subjects) and in oxygen (5 subjects).

Each point represents mean from 8-10 CO_2 tests or 4-5 control tests.

Tidal volume changes were accepted as a sufficient index of ventilatory change; the mean V_T for the reference period was taken over the ½-min before each test; the V_T of the 4th breath was found for each test.

For each subject (one occasion). For oxygen and air runs respectively, the mean ref., and mean 4th-breath values were found and compared by student's t-test, for control, and for CO_2 tests respectively; also the mean 4th-breath values for control tests were compared with mean 4th-breath values for CO_2 tests in any one run. The results of this analysis are shown in Fig. 3:5.

Air: In all subjects the mean 4th breath was larger than the mean ref. breaths. In three of four the difference was significant at at least the 5% level (N, W, S, Fig. 3:5). In two the 4th-breaths in the CO_2 tests were significantly greater than 4th-breaths in control tests (N, W).

Oxygen: In only one of 5 subjects was there any indication of a rise in ventilation after the CO_2 tests: and in this one the response was almost as great as in air (subject W, Fig. 3:5).

Table 3:1

Double breath tests; End-tidal CO_2 levels

Subject	AIR			OXYGEN		
	Reference level		Test rise	Reference level		Test rise
	1st test	last test		1st test	last test	
N	34	34	7	31	30	8.5
K	39	39	6.5	36	34	7
W	40	39	7	38	36	7
S	40	40	6.5	40	34	8
C	-	-		34	36	6

End-tidal PCO_2 to nearest 0.5 torr, at the beginning and end of run of CO_2 and control tests. "Test rise" is the average difference in end-tidal PCO_2 between the breath before the test and the 2nd breath of CO_2 mixture (see Fig.3:3,4).

CO_2: The actual baseline values for end-tidal CO_2 for the several subjects are shown in Table 3:1 The level changed slightly during some experiments and the reference values for the first and for the last test in each sequence are given. The PCO_2 was consistently lower on oxygen than on air. The pre-test end-tidal level, and the end-tidal level of the second breath of CO_2 mixture was found for each test and taken as the stimulating rise of CO_2: the mean rise in any one run is shown in the Table. The range was 6-9 torr.

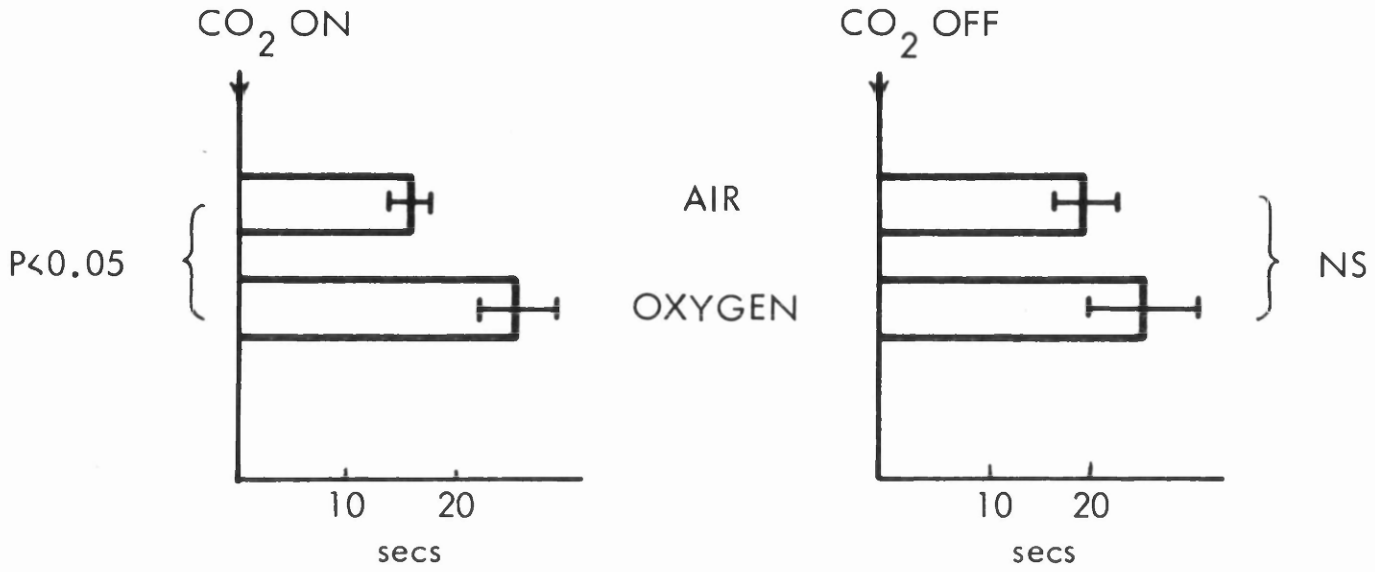
Result of continuing change to CO_2 mixture

The mean times to the first increase in ventilation after the switch on of CO_2, and the mean interval to the first decrease after the switch off, were found and averaged for all observations from both series of experiments (Fig.3:6).

In hyperoxia, the ventilatory response to CO_2 was significantly delayed to 30 sec compared with 18 sec in normoxia: these delay times are compatible with the quoted figures of Gelfand et al (1964) ("fast response starting about 6 sec after the fall in end-tidal PCO_2") when allowances are made for the present timing starting at

Fig 3:6

DELAY TIMES FOR CO_2 ON AND OFF TRANSIENTS



Interval before start of increase (decrease) in ventilation when CO_2 mixture is switched on (off).

for the beginning of the first inspiration of CO_2 mixture, and for the present series being carried out against a background of normal ventilation rather than raised ventilation with hypercapnia. Smaller tidal exchange gives a less rapid change in alveolar gas following a step change in inspired gas. Gelfand's slower components "lag the onset of the first one by about 10 secs": this is confirmed in the timing of our ON-transients.

The timing of the OFF-transients shows less clear differences, and although the mean delay was longer in hyperoxia, the difference did not reach statistical significance, chiefly because of wide variation in the hyperoxic off-transient.

Discussion

Response in normoxia

By recording continuously the end-tidal CO_2 level it has been possible to infer the actual stimulating rise of arterial PCO_2. This was about 8 torr over 2 breaths. The smallest step change studied by Dutton et al (1967) in dogs was of 13 torr PCO_2, and this produced a ventilation ratio response of x2 with a delay of 1-5 sec from direct application by perfusion of the carotid region. The

present changes seen, in air, were of the order of 1.5 ventilation ratio 10-15 sec after a change at the mouth. The change was less rapid in these experiments, and the delay time less accurately assessable, but the values seem compatible. The timing is also similar to the results of Dejours et al (1958) already quoted. In the experiments of Leitner et al (1965b) where impulse traffic and ventilation were recorded, the rise in PCO_2 at the chemoreceptors after a change of inspired gas was taken to about 3-4 torr, although no continuous measurements were made.

These similarities suggest that the responses seen in the experiments with normoxic background were in fact arterial chemoreceptor responses.

Effect of baseline CO_2 level on the response

There was no tendency for a greater response to be associated with a higher initial PCO_2; indeed the subject with the greatest response had the lowest PCO_2 (N: see Fig.3:5 , and Table 3:1).

Response in hyperoxia

The baseline PCO_2 values were consistently lower

than during air-breathing either already by the time of the first test, or by the end of the run. This represents the stimulating effect of oxygen breathing, which will be discussed in the next section.

This meant that the level of PCO_2 reached during the test was lower than in the normoxic tests, so that a strict comparison cannot be made between the two situations, although the rise of PCO_2 was similar. Although in the normoxic series there was no relationship between baseline PCO_2 and extent of response when different subjects were compared, nevertheless within subjects the difference in baseline PCO_2 might well alter the response. It is of interest that the only subject who did show a positive response in hyperoxia, in chemoreflex timing, was the one whose PCO_2 remained continuously nearest to his air-breathing value.

Notes on individual subjects

Subjects N, K and W had been studied in a completely different series of experiments, in the course of which a rebreathing CO_2 response test (in oxygen) was carried out. It is perhaps of interest that subject K who showed the least response in these double breath tests,

also had the consistently lowest response of 8 subjects to the rebreathing test. Also that subject W, who showed a significant response to CO_2 in oxygen in the double breath rests, had the second highest rebreathing response of 8 subjects. Subject N was intermediate in the rebreathing response.

Comparison with Cunningham's method and results

Cunningham et al concluded from the study already referred to, that there is no continuing response of the peripheral chemoreceptors to high CO_2 in the presence of full oxygenation. This was taken to follow from their actual observation:- in a steady state of increased ventilation with raised PCO_2 in hyperoxia, there was no remaining peripheral chemoreceptor activity contributing to the ventilatory drive, because the abrupt and transient removal of CO_2 from the inspired air caused no FALL in ventilation in chemoreflex timing. This does not necessarily imply that a RISE of CO_2 from normal in hyperoxia could not stimulate the peripheral chemoreceptors. In the steady-state and withdrawal situation, the response of the chemoreceptors could have adapted; or, in any situation, the increase of ventilation due to a rise in

CO_2 might not have the same pattern as the decrease of ventilation due to a fall in CO_2. There is evidence for both these possibilities in the findings of Dutton et al (1967): they demonstrated a greater initial, than continuing, increase in ventilation in response to a sustained step increase in CO_2, and they also demonstrated that by contrast there was no undershoot of ventilation but an exponential type of decrease when the CO_2 was abruptly reduced.

Conclusions

The presence of a rapid response of ventilation to a transient increase in PCO_2 in normoxia has been confirmed, but shown to be variable in its extent between subjects.

In hyperoxia, evidence has been presented that in some subjects there may be a rapid response to increase in PCO_2 with a time course which is not distinguishable from that in normoxia, and which is therefore reasonably attributable to peripheral chemoreceptors.

The time delays found at switching to continuing CO_2 mixture show on average a difference between air and oxygen background which supports the concept of a peripheral chemoreflex component active in normoxia but not in hyperoxia.

THE EFFECTS OF BRIEF HYPEROXIA ON VENTILATION AND HEART RATE

Introduction

- Ventilation
- Heart rate
- Present study

Methods

- Subjects
- Procedure
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Results

- Ventilation
  - Initial depression
  - Secondary stimulation
    - Timing and extent
    - Relation to initial PCO_2
  - Discussion of ventilatory changes
    - Measurement of mixed venous PCO_2
- Heart rate
  - Results
  - Discussion

Discussion

Conclusions

THE EFFECTS OF BRIEF HYPEROXIA ON VENTILATION AND HEART RATE

Ventilation

It is now well established that breathing pure oxygen leads, even at rest, to transient depression of ventilation; the extent of this depression has been equated with the peripheral chemoreceptor hypoxic drive, (Dejours, 1963), or more strictly, the hypoxia + CO_2 and/or pH drive (Lambertsen et al, 1963).

Oxygen has also been repeatedly reported to stimulate ventilation later, when the initial depression has passed off. Shock & Soley referred to this phenomenon in 1940, and attributed it to impaired CO_2 transport as a consequence of greater venous oxygen saturation than normal (the C-D-H effect: Christiansen, Douglas & Haldane, 1914). Comroe's group reported stimulation in some subjects, and later published more detailed findings which showed a definite mean increase in ventilation in the 6th-8th min (Watt et al, 1942; Dripps & Comroe, 1947). Lambertsen's group has contributed observations of this increase and comments on its causation (Lambertsen et al, 1953; Lambertsen et al, 1963). There appear, however, not to be detailed reports on the timing

of the increase in ventilation, which might or might not confirm its suggested explanation.

Heart rate

Breathing 100% oxygen is known to lead to bradycardia. Whatever else may happen by way of ventilatory or circulatory changes, this is the most predictable occurrence of all (Ledingham, personal communication).

The following findings refer to normal men:(at 1 atm)

Dripps & Comroe (1947) reported a significant decrease in heart rate during 8 min of oxygen breathing, and equated this with a pure estimate of hypoxic chemoreceptor drive since,unlike the ventilatory response, there was not, for heart rate, any secondary opposing effect comparable to an increase in PCO_2.

Daly & Bondurant (1962) reported the circulatory effects of oxygen breathing: cardiac output decreased, entirely on account of bradycardia; mean arterial blood pressure increased: there was an increase in peripheral vascular resistance. All the effects were abolished by atropine, but the response still occurred during reactive hyperaemia; inspired oxygen concentrations less than 100% were also used, and the heart rate

decrease related linearly to the PO_2. They concluded that oxygen depressed chemoreceptors, progressively from 100% inspired downwards, producing reflex bradycardia by vagal stimulation, and that the vasoconstriction was secondary: conclusions which do not appear to be justified from their findings. Their observations are comparable to those of other workers.

Other evidence for the vasoconstrictive effect of oxygen, from limb blood flow measurements, was presented by Bird & Telfer (1965) and also by Pollock, Bell & Ledingham (1968) who showed a decrease in muscle blood flow, increase in blood pressure, and decrease in heart rate; the changes were established at the time of the first measurement about 5-10 min after the start of 100% oxygen breathing at 2 atmospheres.

Dejours et al (1958) reported that a reduction in heart rate following a single-breath oxygen-test occurred in resting subjects slightly later than the depression of ventilation (p 519) Like the ventilatory depression, the effect was distinguishable against a normoxic background and was greater against a hypoxic

one (p.1/14). There is no other reported study on the timing of the depression or on the relationship of that timing to the other cardiovascular responses.

Present study

The object of the present study was to define the incidence, extent and timing of the secondary stimulation of ventilation which may occur in the course of a few minutes breathing pure oxygen, and in some later experiments, to make similarly detailed observations on heart rate.

Methods

The general instrumentation was as already described.

Bags of air and of oxygen were attached to the sliding valve; ventilation was monitored by inspiratory pneumotachograph and by continuous analysis for CO_2 by sampling at the mouth; in some experiments heart rate was monitored by ECG, triggering a Devices ratemeter, so that beat-by-beat heart rate was recorded. (see p. 5/25).

Subjects

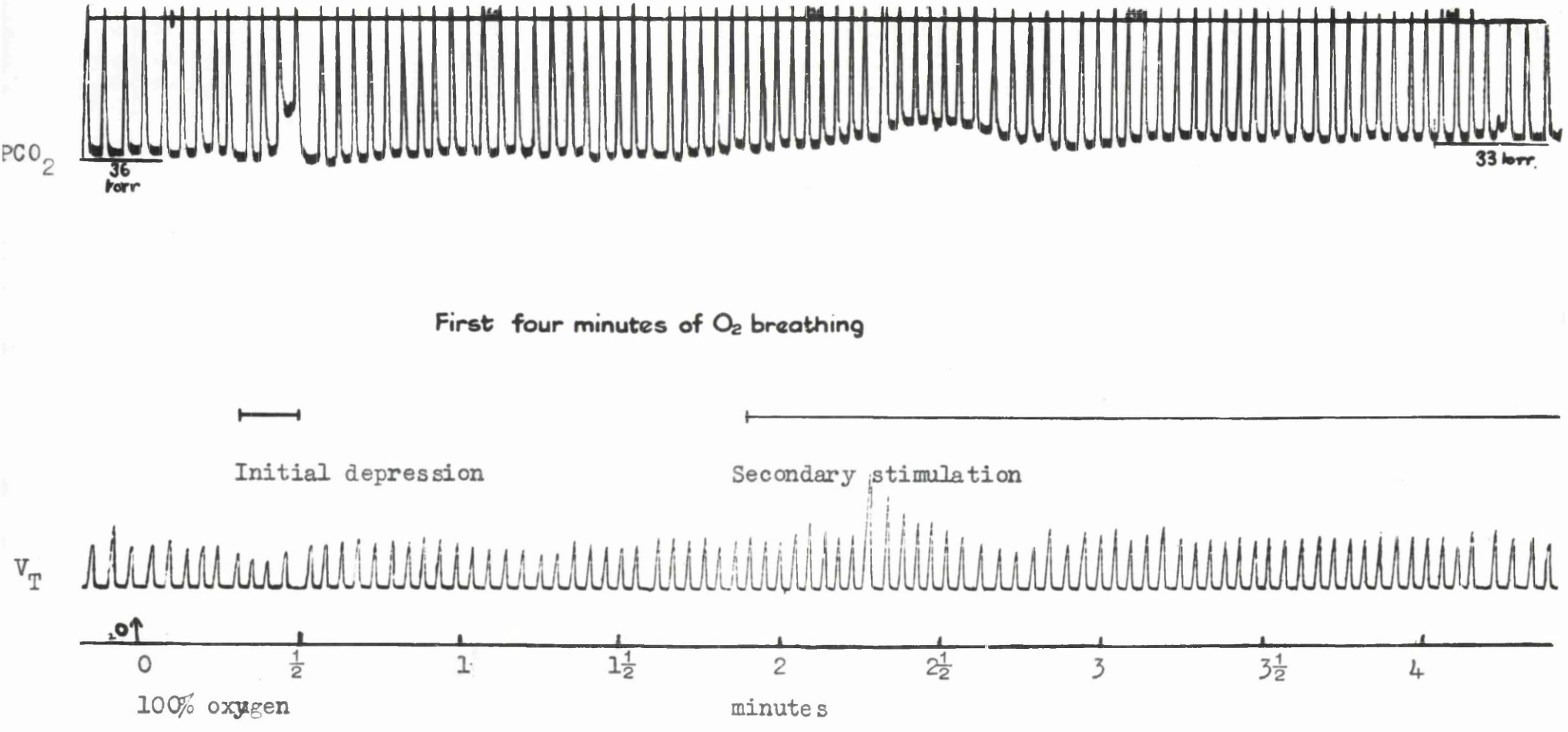
These were all normal young men. Some of the data were taken from a series of control experiments for

a drug trial, in the course of which a 2 min period breathing oxygen was routinely applied as a preliminary to a rebreathing CO_2 response test (Jennett, 1968). This comprised 8 subjects, each studied at least 4 times. In addition to this three subjects were studied in association with the double-breath CO_2 tests reported in the last section, or with the hypoxic series reported in the following section. On some occasions, the air → oxygen change, with a control air → air change, was studied alone; on some it preceded another type of test, on some followed it: where several tests were performed on a single occasion the order was randomly determined, and ventilation and heart rate levels allowed to return to baseline between all tests. Subjects were studied semi-recumbent.

Procedure

The subject's heart rate and ventilation were allowed to settle to steady levels whilst he was breathing room air through the apparatus. Air from a bag was then inhaled. A sham change was made by switching to air from another bag, and this was maintained for 2 min, then a switch to 100% oxygen was made. In some experiments this was maintained for 2 min only; in others it was

Fig. 4:1


PCO2
36 torr
33 torr.
First four minutes of O2 breathing
Initial depression
Secondary stimulation
VT
0
½
1
1½
2
2½
3
3½
4
100% oxygen
minutes

allowed to continue for 10 minutes.

Extraction of data

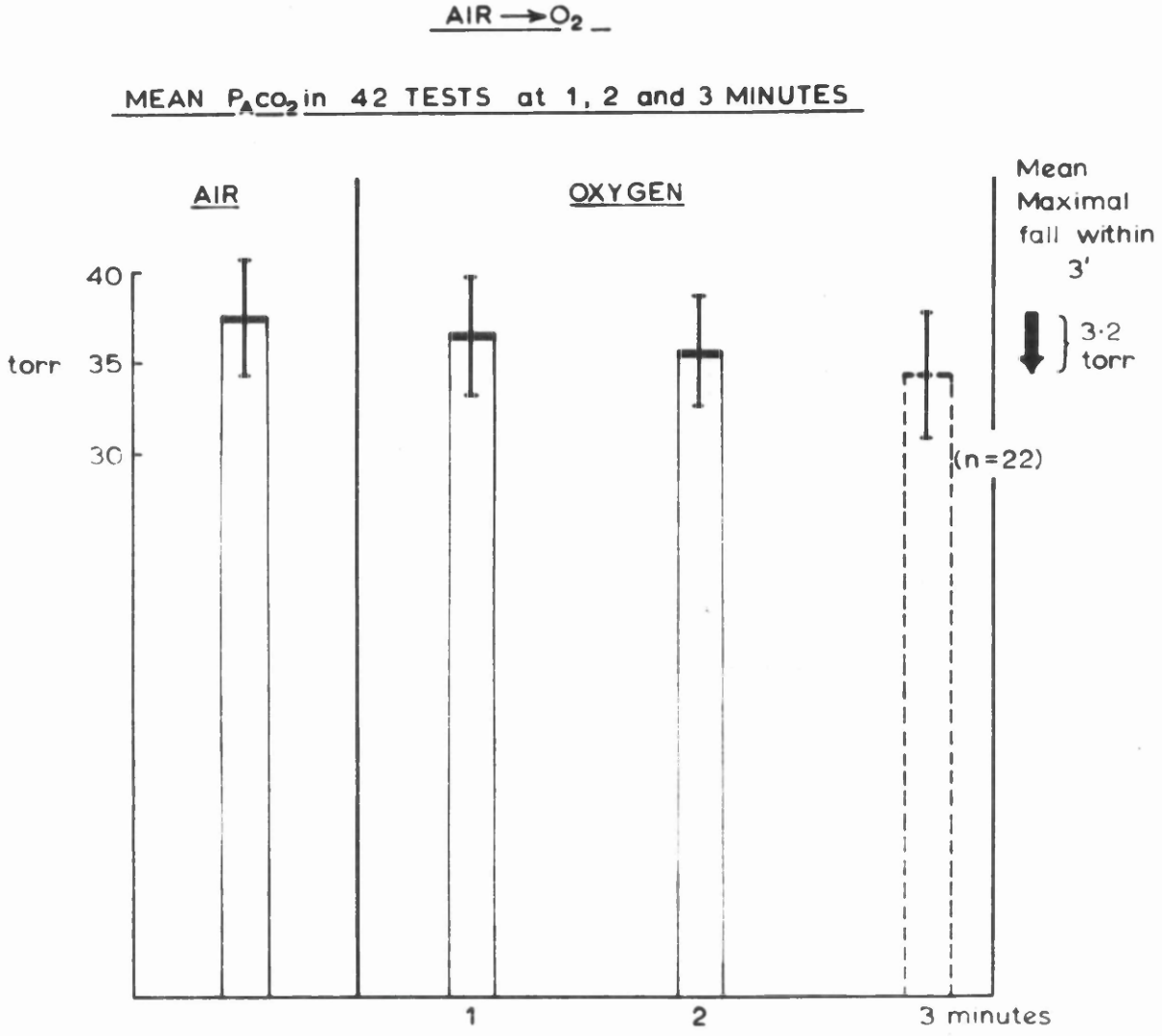
1) Initial changes: The tidal volume and end-tidal PCO_2 were found for a reference period of one min before the switch to oxygen, and for each of the first 8 breaths on oxygen.

2) Later changes: The end-tidal CO_2 (mean over several consecutive breaths) was found at $\frac{1}{2}$ min and 2 min; and where available at 8 min and 10 min. When heart rate was recorded, the rate was found for the periods of the first and second half minutes on oxygen, and for each succeeding minute.

Results

Fig. 4:1 shows an example of an experimental record, showing both initial depression and secondary stimulation of ventilation. 63 changes from air to oxygen were examined for the occurrence of initial ventilatory depression, in the first 10 to 20 sec. In this series a depression was taken to be present if one or more breaths in the appropriate timing showed an end-tidal PCO_2 at least 2 torr higher than the reference level. By this criterion there were only 8 instances of depression.

Fig. 4:2



Mean changes in P_ACO_2 during the first 3 minutes of breathing 100% oxygen, in those experiments where a secondary stimulation of ventilation was seen.

The bars represent the standard deviation of each observation

The maximal decrease in PCO_2 sometimes occurred in the second or third minute. The mean maximal fall is therefore given separately irrespective of the time at which it occurred.

These observations were not the main concern; for timing they serve to confirm the findings of other workers: they suggest, however, that this depression may vary considerably between individuals.

Secondary stimulation

77 changes from air to oxygen were examined for the occurrence of increased ventilation during the first 2 or more minutes, and this was found in 52 (67.5%). The criterion of stimulation was a continuing (progressive, or maintained) decrease in end-tidal PCO_2 of 2 torr or more. Such a change was not seen in any of the control air ⟶ air changes.

Effect and extent of stimulation

When stimulation occurred, it most commonly started between 1 and 2 min after the first breath of oxygen (55% of instances); earlier in 35% and later in the rest.

The decrease in PCO_2 usually appeared to be still continuing at 2 min in those experiments in which this was the duration of O_2 breathing; in those continued to 3 or 4 min there was a further decrease (Fig. 4:2); in those followed to 10 min, there was no further

Fig 4:3

RELATIONSHIP BETWEEN INITIAL P_Aco_2 AND: —

1. Occurrence of Stimulation

All Tests (63)	
Stim.	Not Stim.
(42)	(21)
37.5 (S. D. 3.1)	36.1 (S. D. 2.1)

Individuals			
		Stim.	Not Stim.
Subject	1.	40.9	39
	2.	(30.2	32.2)*
	3.	39.8	36
	4.	39.3	36
	5.	38.1	36
	6.	40	37
	7.	39	37

(*only subject who had lower value for PCO_2 when stimulation occurred)

2. Time of Onset of Stimulation

(42) Time of onset	< 1 min (15)	1-2 min (23)	> 2 min (4)
Initial P_Aco_2	37 (S. D. 2.5)	37.8 (S. D. 3.5)	37.5 (S. D. 3.7)

(Results from 63 test, 42 in which stimulation of ventilation occurred during the first 2-3 min of 100% oxygen breathing (Stim.) and 21 in which it did not (Not Stim.)

significant change.

Occurrence of stimulation in relation to initial PCO_2

If the stimulus to ventilation were dependent on a tendency for central tissue PCO_2 to increase, then there might be a longer delay before an appropriate level for stimulation were reached if the initial P_ACO_2 were lower. However, a low P_ACO_2 would be accompanied by a reduction in cerebral blood flow probably sufficient to keep cerebral tissue PCO_2 normal: thus the initial P_ACO_2 might make no difference. The results were considered from this point of view: there was in fact no correlation between occurrence of stimulation and level of initial P_ACO_2 between subjects; within subjects there was a suggestion of a greater tendency to stimulation with an initially higher P_ACO_2. This is probably not of any importance (Fig. 4:3).

Discussion of the ventilatory changes

Validity of methods

Due allowance was made for altered carrier gas in the CO_2 analysis; any effect due to this would, in any case, have been equally evident in all instances and

subjects. The effect of substituting oxygen for air is to give a very slightly higher reading for the same actual CO_2%. To neglect this would therefore be to exaggerate the initial depression, and underestimate the secondary stimulation. In a trace an apparent decrease of PCO_2, as shown in Fig. 4:1, is therefore at least as real as it looks.

The change to oxygen also affects the pneumotachograph reading (p. 2/3). A correction was made for this; the effect of ignoring it would be to underestimate the initial depression, and exaggerate the secondary stimulation. But the change would be evident, and constant, from the first breath.

Although in this series ventilation was not measured from the pneumotachograph recording, but changes assessed from the resulting P_ACO_2, the tidal trace was always examined to make sure that it changed consistently with the P_ACO_2 changes.

Significance of results

Similar increases in ventilation have been reported during pure oxygen breathing, and attributed to

an increase in tissue PCO_2: when haemoglobin becomes less desaturated than usual in the tissues, a greater PCO_2 difference between cells and capillaries would be necessary to maintain the rate of removal of CO_2. This would mean that ventilation would be stimulated as soon as arterial blood became sufficiently oxygenated to make an appreciable difference to desaturation in the cerebral capillaries: the tissue PCO_2 would increase until or unless ventilation increased, reducing the arterial PCO_2 and widening the A-V difference, restoring tissue PCO_2 to normal. A new steady state can be envisaged at a higher ventilation, and lower CBF. The predicted change would be very small, however, at 1 atmosphere oxygen.

This explanation is supported by Lambertsen's results for arterial and jugular PCO_2 in men, during air- and oxygen-breathing respectively.

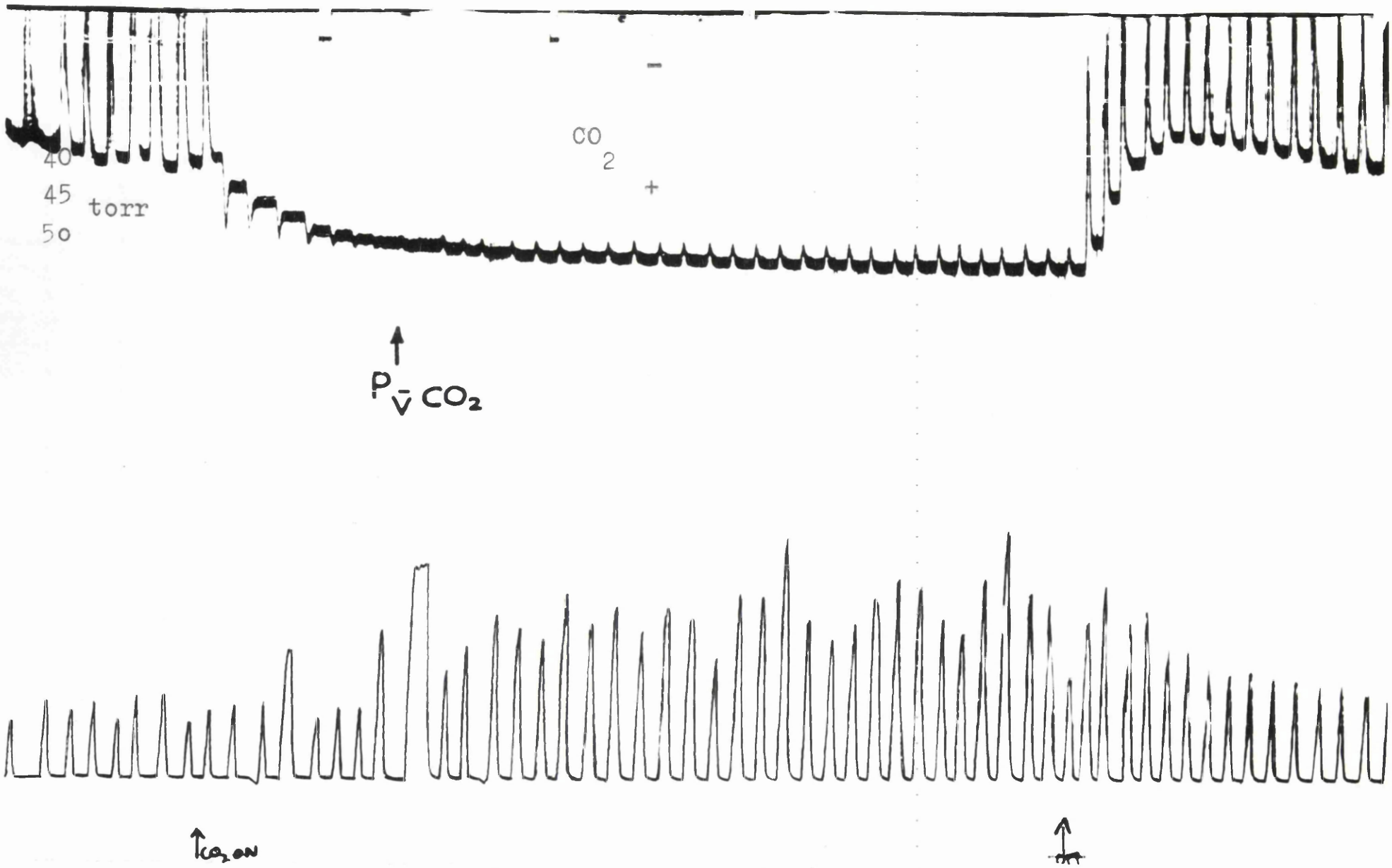
> The A-V difference was greater - P_ACO_2 was lower, and jugular PCO_2 higher, by about 2 torr. These results were from 4 subjects only, but represent some of the very few available at 1 atmosphere of oxygen (Lambertsen et al, 1963). The P_ACO_2 values are comparable to the present results. Meyer, Gotoh & Takagi (1967) measured various parameters continuously, including $P_{jv}CO_2$, during inhalation of different gas mixtures, including 100% oxygen. The one published trace for 100% O_2 shows a rise in $P_{jv}CO_2$ of about 3 torr occurring in the second

minute: but they also show a slight increase in P_ACO_2. The timing of the change would fit in with the present results.
Dripps & Comroe (1947) measured the ventilation in the 6th-8th minute on 100% oxygen, and showed that it had increased on average by about 8% above control level. This would represent an approximate reduction of alveolar PCO_2 of 3 torr; therefore the present results are comparable.
In discussion, these authors prefer to ascribe the stimulation to some lower respiratory tract reflex induced by 100% oxygen, rather than to the C-D-H effect or reduced CBF: their grounds are that Kety told them there was no significant rise in jugular venous PCO_2 during oxygen breathing.

The timing of the stimulation in the present series is compatible with the time it would take before the cerebral vascular bed became fully oxygenated; it is appropriately longer than the timing of the initial depression, which acts at the chemoreceptors when the P_aO_2 becomes, say, over 120 torr - a point which would be reached several breaths before full oxygenation at around 650 torr.

The question next arose as to whether any other observations could be made over the same early minutes of oxygen breathing, using the equipment available, which might support this explanation of ventilatory stimulation:

Fig 4:4



Measurement of mixed venous PCO_2 by rebreathing test.

i) If the C-D-H effect has developed at the time when ventilation increases, it might be reflected also in a rise in mixed venous PCO_2.

ii) The depression of heart rate also might be related to the development of tissue acidity, or alternatively to the effect of full oxygenation in producing increased peripheral resistance: so the timing of heart rate changes might be similar to that of ventilatory stimulation.

Mixed venous PCO_2 measurement

The $P_{\bar{v}}CO_2$ was estimated during air breathing and after 4 minutes of oxygen breathing in 11 subjects.

Method

The estimation was similar to the method of Campbell & Howell (1962) but with continuous recording of CO_2.

The subject was switched into a rebreathing bag containing oxygen; he rebreathed for 1½ minutes, then was switched back to air (or oxygen) for a 2-minute interval; he then rebreathed from the same bag again, for about 40 seconds. PCO_2 at the mouth was recorded throughout, so that it was possible to identify a plateau

TABLE 4:1

Values for mixed venous PCO_2

	AIR			OXYGEN		
Subject	P_ACO_2	$P_{\bar{V}}CO_2$	$\bar{V}$-A	P_ACO_2	$P_{\bar{V}}CO_2$	$\bar{V}$-A
1	41	49	8	39	44	5
2	36	42	6	31	42	11
3	38	43	5	37	48	11
4	39	45	6	34	50	16
5	36	47	11	31	45	6
6	35	44	9	35	46	11
7	30	34	4	33	38	5
8	35	41	6	35	47	12
9	34	40	6	31	39	8
10	35	41	6	37	45	8
11	40	45	5	40	45	5
Mean	36.3	42.8	6.55	34.8	44.45	8.91
S.E.	0.935	1.2	0.61	0.96	1.09	1.08
Significance of difference, Air v. O_2:			N.S.		N.S.	P=0.1 - 0.05

level during the second rebreathing, which corresponded to mixed venous PCO_2. When such a level was not reached, no value could be estimated. In later runs, the first rebreathing was dispensed with by using a bag of 7% CO_2 in oxygen initially. Similarly it was possible to identify an equilibration level as $P_{\bar{v}}CO_2$, on the continuous trace (Fig. 4:4). This test was carried out (a) during airbreathing and (b) after 4 minutes oxygen breathing, the order of tests being determined randomly. The test itself demands a period of oxygen breathing, and is said to estimate the "oxygenated mixed venous PCO_2"; however the equilibration point is reached during the first 20 seconds of the oxygen breathing. Therefore, even with the higher than normal ventilation due to high CO_2, the time is probably too short for full development of the C-D-H effect. It therefore seems valid to compare these $P_{\bar{v}}CO_2$ values as representing the levels when breathing air and oxygen respectively.

Results

The results are shown in Table 4:1 The difference between air and oxygen values for $P_{\bar{v}}CO_2$ were not statistically significant (by comparison of means by t-test)

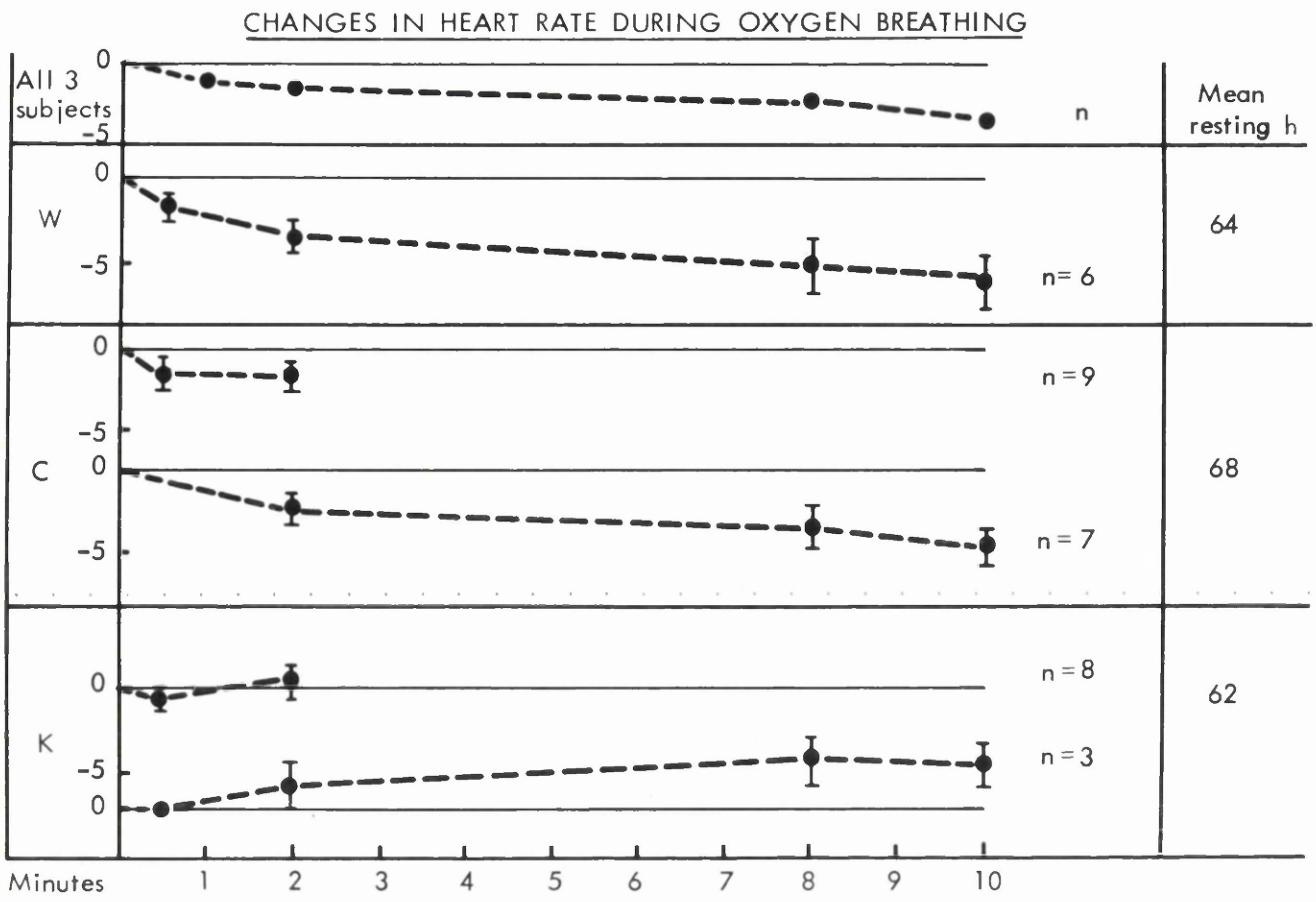

CHANGES IN HEART RATE DURING OXYGEN BREATHING
All 3 subjects
0
-5
n
Mean resting h
W
0
-5
n= 6
64
C
0
-5
n = 9
0
-5
n = 7
68
K
0
-5
0
n = 8
n = 3
62
Minutes
1
2
3
4
5
6
7
8
9
10

Fig 4:5

but there was a higher mean value during oxygen breathing. The increase in the A-V difference for PCO_2 during oxygen breathing fell just short of significance at the 5% level.

Results of heart rate measurements

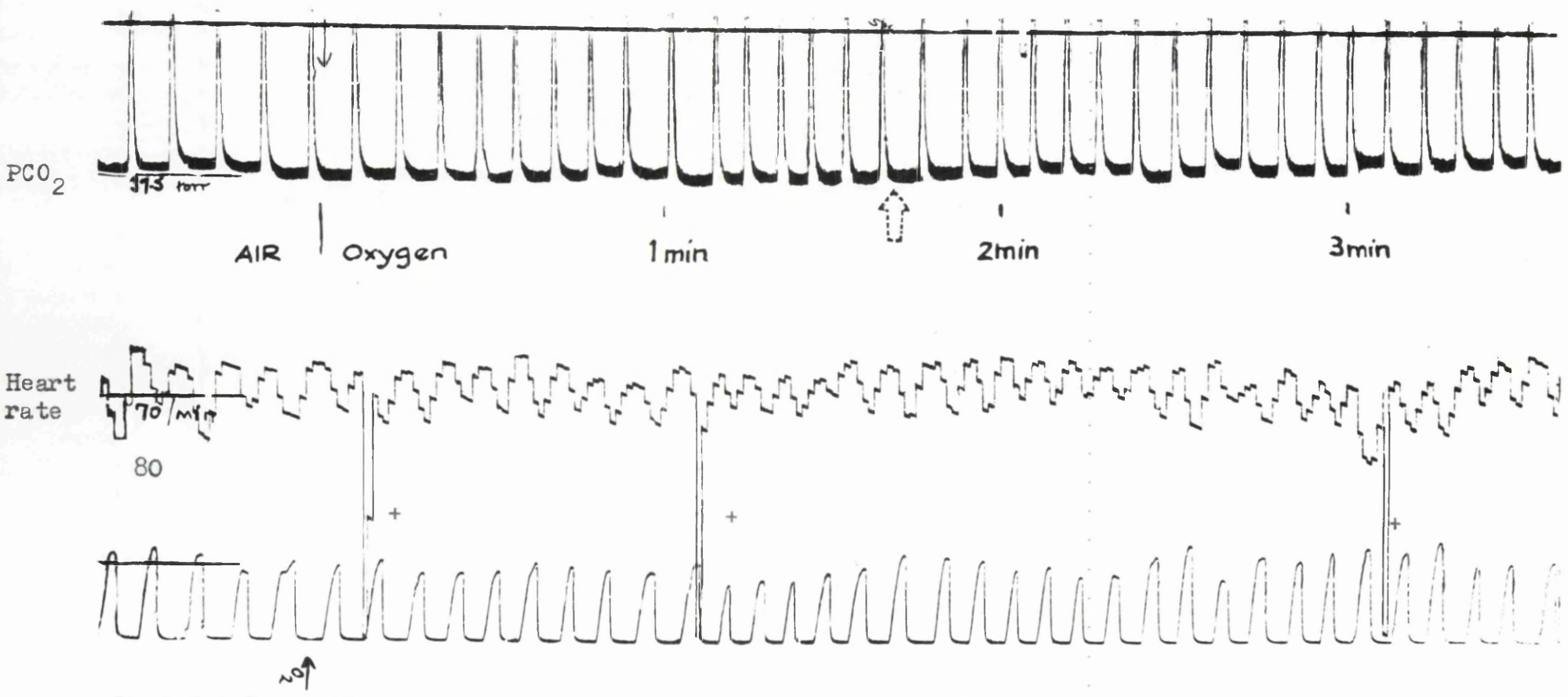
In 33 experimental runs, on 3 subjects, the heart rate was determined for two minutes of oxygen breathing. The mean changes for each subject, and the overall means for all runs are shown in Fig.4:5 Two subjects consistently developed bradycardia: the third did not. In 16 runs the heart rate was followed for 10 min of oxygen breathing; in the 2 subjects, the decrease continued; in the third there was still no tendency to decrease. All records were examined for indication of a decrease in heart rate in the first 20 seconds, which could be attributable to chemoreceptor effect or to a reduction pari passu with reduced ventilation. In some runs a reduced sinus arrhythmia amplitude occurred with the ventilatory depression. Otherwise there was no heart rate change comparable to the depression of $\dot{V}$ in timing or extent.

Discussion of the heart rate changes

The changes found in the first ten minutes of oxygen breathing were minimal; however the overall

Fig 4:6

Effect of 100% oxygen breathing on ventilation and heart rate in the first $3\frac{1}{2}$ minutes.



No initial ventilatory depression is seen in this experiment (compare Fig. 4:1)

Arrow indicates time at which ventilation starts to increase, PCO_2 to decrease.

There is no sustained alteration of heart rate (+artefacts)

impression was of a slowly developing change, starting within 2 minutes and progressing very slightly in succeeding minutes. The reduction at 10 min is less than that quoted by Daly & Bondurant (they do not give heart rate figures as such, but from their published values for cardiac index, which they said changed entirely due to heart rate, a reduction of about 9-10 beats per min can be calculated). The change could be taken, on the grounds of timing, to reflect the same chemical occurrences as the secondary stimulation of ventilation. There is no evidence to support the cancellation of a chemoreceptor hypoxic drive as the cause of bradycardia, because there was no step change in a chemoreceptor timing; if there was a slight reduction pari passu with the ventilation, the heart rate recovered, and then decreased gradually again. The results are comparable to those of Dripps & Comroe (1947) who reported an average, significant, decrease of -4.5 beats per min at 6-8 min after the start of oxygen breathing. They did, however, also find a significant depression of heart rate in the first 2 minutes, when the lowest value in this period was compared with the lowest value in the preceding minute breathing air.

It would be of interest to know the time course

in similar circumstances of:

(i) the changes in cerebral blood flow: It is acknowledged that there is a reduction in cerebral blood flow in man in hyperoxia. Lambertsen associates this with the reduction in arterial PCO_2 due to the ventilatory stimulation. If, however, the ventilatory stimulation is thought to be due to increased tissue PCO_2, and brain blood flow is thought to be dependent on tissue PCO_2 or acidity rather than on arterial PCO_2 itself (Severinghaus et al, 1966), the CBF should be normal or increased. One can imagine that a new steady state is reached in hyperoxia where a balance between a reduced CBF, an increased ventilation, and the C-D-H effect maintain extracellular pH at the normal value: but the causes and the effects, the sequence of stimuli and responses in the attainment of this state are not obvious.

(ii) changes in peripheral vascular resistance: Daly & Bondurant reported increased blood pressure and peripheral resistance, which they appear to attribute to a decrease in heart rate. It could be that vasoconstriction and rising blood pressure caused the bradycardia. Further studies incorporating continuous blood pressure measurement

would be required to settle this point. Published reports of experiments where blood flow BP, and pulse rate were measured, include no continuous measurements in the first minutes.

The gradual development of the bradycardia, rather more prolonged than the development of ventilatory stimulation favours haemodynamic changes as the cause.

It is hoped eventually to repeat studies similar to these with the addition of the following refinements:

1) continuous monitoring of end-tidal PO_2
2) continuous blood pressure recording
3) frequently repeated measurement of forearm blood flow by strain gauge plethysmography.

Summary and conclusions

The extent and timing of the changes in ventilation and in heart rate in the first minutes of 100% oxygen breathing have been observed. Ventilation increased during the second and third minutes, which is compatible with minimally increased cerebral tissue PCO_2 as the stimulus.

Heart rate decreased very slightly and very gradually on average over the course of 10 minutes, and

was not significantly altered at 2 minutes. This is not compatible with a cancellation of chemoreceptor drive, but suggestive of bradycardia developing as a result of increased peripheral vascular resistance.

Summary of possible causes of tachycardia

4. Methods of study applicable to man

i) Timing of responses in normal subjects

Literature

Approach to present studies

ii) Investigation of subjects with pathological destruction of some component of the possible mechanisms causing tachycardia.

CARDIOVASCULAR AND VENTILATORY RESPONSES TO HYPOXIA, AND THEIR RELATION TO EACH OTHER

Compensatory mechanisms in the oxygen delivery system

Gaseous hypoxia can be compensated by an increase in one or both components of the oxygen delivery system - ventilatory and cardiovascular. An increase in the one needs to complemented by an increase in the other, yet to a certain extent they are alternatives: the higher the PO_2 can be raised by ventilation, the less does cardiac output need to increase to maintain the whole-body oxygen flow and the average tissue oxygen tension. The secondary effect of ventilation, however, complicates this; changes in PCO_2 affect blood flow, particularly in the brain; the gain in blood oxygen content is offset by the reduction in blood flow.

The balance between the two responses varies considerably between species: rabbits, on the one hand, increase their ventilation steeply in hypoxia, and their cardiac output often not at all (Korner, 1959) whereas man increases his cardiac output considerably and constantly, and his ventilation relatively little. Dogs, on which most experimental observations have been made, show both responses.

These species differences, and the possibility also of altered responses during anaesthesia, make results of animal experiments of limited relevance to man.

In man there is less direct information on cardiovascular responses than there is on ventilatory response to hypoxia: with a few exceptions (Dripps & Comroe, 1947; Reed & Kellogg, 1960) the two are unfortunately seldom studied simultaneously.

Ventilatory response to hypoxia

This was discussed as part of the general introduction (p1/9-15) and only the essential points will be restated, with particular reference to man.

Hypoxaemia is a stimulus to ventilation, but only notably so when it is severe. The mechanisms are such that increased ventilation occurs only at PO_2 levels when it will usefully increase the oxygen content of the arterial blood.

The carotid and aortic bodies are the only receptors responsible for the increase in ventilation in hypoxaemia. They do not respond to tissue, or anaemic,

hypoxia (Comroe & Schmidt, 1938) nor does this type of oxygen lack provoke any ventilatory response. When gaseous hypoxia has increased the ventilation, additional tissue hypoxia does not increase it further (Harrison & Blalock, 1927 - dogs made anaemic; Asmussen & Chiodi, 1941 - man, exercising with carboxyhaemoglobinaemia).

There are interactions both between hypoxia and other chemical stimuli at the chemoreceptors, and between incoming chemoreceptor impulses and other stimuli at the respiratory centre. Also there is a direct depressant effect of hypoxia on the respiratory centre. The overall effect of hypoxia is therefore complex.

There are two approaches to understanding the overall ventilatory response to hypoxia; experiments such as those of Riedstra (1963), which examine the responses of the component parts, can lead to reconstructive descriptions, and to models representing interactions (Torrance, 1968); and the whole animal, notably man, has been exposed to hypoxia in many different steady state conditions, and the response of ventilation quantitatively analysed; but a "particular representation is a purely arbitrary one which refuses to break the seal of Pandora's Black Box" (Grodins, 1964).

The method of investigating the response of whole man to hypoxia which is of most interest in the present context, is that which depends on the measurement and timing of immediate responses, before secondary effects have had time to occur; this has been utilised extensively by Dejours and his group, who have thereby established the existence of a small hypoxic drive in man at rest, They have also applied the method to defining the extent of the hypoxic drive at different levels of hypoxia and activity.

Cardiovascular response to hypoxia

It seems from the literature that the tachycardia which occurs in most animals in response to hypoxia, whether of the hypoxic or the anaemic type, is a very constant and specific phenomenon. Yet in the reviews and symposia on cardiovascular effects the conclusions are far from clear-cut: the general impression is one of wide species differences and of incompatibilities between different observations on the same species, perhaps accounted for by different experimental conditions (Korner, 1959; Hatcher & Jennings, 1966). Korner summarises: "the cardiac response is fairly non-specific and is chiefly related to the pattern of peripheral vasomotor tone."

In brief, findings in animals and man are as follows:

In most hypoxic situations studied, heart rate increases. This is usually associated with an increase in cardiac output, and usually largely accounts for it, though stroke volume may also increase. The rise in cardiac output is associated in the pulmonary circulation with a disproportionate increase in pulmonary artery pressure due to increased vascular resistance, most probably in the veins; in the systemic circulation, the effect varies between species, depending on the relative preponderance of sympathetic vasoconstriction, or of local vasodilatation, particularly in skeletal muscle. In dogs there is characteristically a hypertension: in man there is usually no change in arterial blood pressure unless hypoxia is severe.

RESPONSES IN AWAKE MAN

The tachycardia of hypoxia has been well-known in man at least since Lutz& Schneider described it in 1919 as developing about 15 sec after starting to inhale pure nitrogen, and Barcroft observed it at high altitude in 1925.

The familiar increase in heart rate occurs even in very mild hypoxia and is inversely related to the oxygen level (Dripps & Comroe, 1947). It is related to the P_aO_2 whatever the accompanying change in P_aCO_2 or in the mechanical level of ventilation (Richardson et al, 1966). It is of the order of an increase of 25% for a P_aO_2 of 35-40 torr, or 8% of inspired oxygen, and the response is discernible when oxygen is reduced in inspired air to 18%.

Unlike the ventilatory response, the tachycardia is also induced by anaemic hypoxia. The subjects partly poisoned by Asmussen & Chiodi (1941) and already referred to as having no increased ventilation attributable to carboxyhemoglobinaemia showed an increase in heart rate which was not associated with any other of the cardio-vascular changes of hypoxaemia.

Cardiac output usually increases, and this has often been said to be largely accounted for by the increased heart rate (Vogel & Harris, 1967); however, a significant increase in stroke volume has also been reported by many workers (Richardson, Kontos, Raper & Patterson, 1967; Harris, Bishop & Segel, 1961).

These authors also confirmed many earlier reports that there is no significant change in arterial blood pressure.

Pulmonary vascular resistance was shown to increase in man by Motley, Cournand, Werko, Himmelstein & Dresdale (1947), although they differed from other authors in finding no increase in cardiac output. Other studies have consistently confirmed the rise in pulmonary artery pressure (Harris et al, 1961; Doyle, Wilson & Warren, 1952). There is some disagreement about the level of hypoxia at which this effect is seen - Korner states that in man it does not occur above a PO_2 of 35 torr; however, Doyle et al gave their subjects 10% oxygen, recorded saturations of 65-75% and showed a doubling of normal pulmonary artery pressure.

Evidence for decreased peripheral vascular resistance was reported by Abramson, Landt & Benjamin (1953); the increase in muscle blood flow has been said to be due to accompanying hypocapnia (Black & Roddie, 1958) but this impression was corrected by Richardson et al (1966) who found the flow to be similar at a given P_aO_2 whether or not PCO_2 was controlled.

Cerebral blood flow increases in hypoxia, although this increase is counteracted by the fall in P_aCO_2 if the ventilatory response is significant; the change is of the order of +35% increase, breathing 10% oxygen (Kety & Schmidt, 1948).

Evidence concerning the mechanism for the increase in heart rate

The tachycardia in hypoxia appears to be sufficiently independent of other cardiovascular changes to justify its separate consideration. The heart rate increases whether or not there is hypertension, and whether or not there is an increase in cardiac output. The causation of the tachycardia is agreed by most authors to be surprisingly obscure.

Ultimately the action must be on the SA node; the evidence will be considered working 'backwards' from this to efferent pathways, possible reflex centres, and possible afferent pathways and receptors.

1) Direct effects on SA node

a) of low PO_2 itself.

This explanation would account for the occurrence of tachycardia in anaemic as well as in hypoxic hypoxia; it has been suggested (Heymans & Neil, 1958) but there is no clear evidence for it.

Evidence from the study of denervated or isolated hearts is inconclusive.

Resnik (1925) observed hypoxic tachycardia in vagotomised dogs: he also removed the stellate ganglia in one dog and still found a response to hypoxia.

Glick, Plauth & Braunwald (1964) used awake dogs, some intact, some with totally denervated hearts; they showed similar tachycardia in response to hypoxia in both groups. On the other hand, Greenfield & Ebert (1963) found a much reduced heart rate response after denervation.

Cross, Rieben, Barron & Salisbury (1963) studied many preparations, including spontaneously beating isolated dogs' hearts: in these they found no change in heart rate when the coronary perfusate was made hypoxic, whilst other factors were kept constant.

b) of haemodynamic consequences of hypoxia

If there is indeed a Bainbridge effect, and if it can be brought about by mechanical stretch of the nodal region, then tachycardia might result from increased venous return or increased right heart pressures secondary to pulmonary vaso-constriction. If this were the explanation, it would be difficult to explain the consistency of tachycardia in relation to the variability of vasomotor responses.

2) Hormonal effects on SA node.

Increased adrenal medullary secretion is an established accompaniment of severe hypoxia, and has often been invoked as the cause of the tachycardia. Hatcher & Jennings (1966) have presented good evidence that in anaesthetised dogs, hypoxic tachycardia is due to a humoral agent which is not, or not only, adrenaline, and which is dependent for its release on (i) innervated adrenals and (ii) the kidneys, not

necessarily innervated. They had evidence of a similarly acting, though not necessarily identical "cardiotonic agent" in anaemia. (The assay method, on a normally oxygenated dog, would not distinguish direct humoral effects on the heart from those at other sites).

In man, plasma catecholamines have been measured during moderate hypoxia, sufficient to cause a significant increase in heart rate, and no increase has been found (Richardson et al, 1967).

3) Neural effects on SA node

(i) The efferent pathway

Either, or both, reduced vagal activity or increased sympathetic activity could account for the tachycardia in hypoxia; the vagal effect was reported to be the main, but not the only one, in 1925 (Sands & DeGraff). Later studies have mostly favoured the sympathetic.

Sympathetic component: In cats Downing (1966) reported a marked increase in impulse traffic in the cardiac sympathetic nerves in hypoxia, despite a concurrent increase in arterial blood pressure (which when induced otherwise could be shown to decrease the impulse traffic).

In man, the effect of hypoxia has been compared before and after interference with the sympathetic pathway by drugs.

a) Beta-adrenergic blockade with pronethalol, or propanolol was found to modify the effect of hypoxia (Richardson et al, 1967). The cardiac output still increased after the drugs but to a lesser extent. When, however, the published results are analysed so as to separate the heart rate from the stroke volume components, it can be shown that the increase in cardiac output which remained after blockade was entirely due to an increase in heart rate: the drug appeared to prevent only the increase in stroke volume. The authors did not make this distinction).

b) Harris et al (1961) found the heart rate response to be unchanged by administration of guanethidine, and concluded that it was unlikely that the sympathetic pathways played an important part in circulatory adjustments to hypoxia.

The evidence, then, is conflicting.

The vagal component: It has been reported (Sands & DeGraff, 1925) that vagal section diminishes and delays the increase in heart rate during progressive hypoxia in dogs. However, this does not necessarily imply that vagal inhibition is the normal mechanism; the initially high heart rate might preclude a further rise, or vagal section might interrupt afferent pathways from the lungs which would lead to tachycardia secondary to increased lung expansion (Daly & Scott, 1962).

There appears to be no direct evidence of decreased vagal activity during systemic hypoxia.

ii) Effect of hypoxia on the central nervous system

Hypoxia could depress vagal centres or stimulate sympathetic centres either directly, or by means of a change in afferent impulse traffic from receptors.

Direct stimulation of sympathetic centres

This is the most widely accepted explanation of the tachycardia in hypoxia.

Hypoxia of the brain: In dogs, separate perfusion with hypoxic blood in the brachio-cephalic distribution has been shown to bring about the normal cardiac response to hypoxia, whereas perfusion of the carotid sinus region alone, causes bradycardia; brain hypoxia caused increase in the activity in the cardiac sympathetic nerves; carotid sinus hypoxia did not alter this activity. Thus Downing's group localised the site of stimulant action to the CNS (Downing, Remensnyder & Mitchell, 1962, and Downing, Mitchell & Wallace, 1963).

The degree of hypoxia in these experiments was very severe (6-30 torr PO_2), and the heart rate increase relatively small: these results cannot therefore be taken to explain satisfactorily the similar tachycardia which occurs in very much milder hypoxia in the intact animal.

Hypoxia of the spinal cord: Mathison (1910) found an increase of spinal sympathetic activity in asphyxia in spinal cats: but tachycardia still occurred when the upper thoracic cord was removed. Since then the possibility of direct stimulation of the spinal sympathetic centres seems not to have been considered except by Alexander (1945) and his findings seem largely to have been overlooked, and unconfirmed. He demonstrated that the deafferented isolated thoracic spinal cord, in cats, gave rise to an increase in activity in the cardiac sympathetic nerves when tissue hypoxia was induced either by inhalation of nitrogen or by reduced blood flow.

Neurones and PO_2: Recently Chalazonitis (1969) has shown that lowering the PO_2 can lower the membrane potential in the giant neurones of snails, and that functionally different neurones may be affected differently by changes in gas tensions and pH. Such evidence supports the possibility of a stimulating effect on some medullary neurones, whilst others (respiratory) are depressed.

iii) Afferent pathways; receptors

a) arterial chemoreceptors

These were taken to be the source of cardiovascular, as of respiratory, chemoreflexes by several authors, although as long ago as 1931 it was demonstrated (by Heymans, Bouckaert & Dautrebande) that drugs known to stimulate the carotid body chemoreceptors gave rise to bradycardia. Bernthal and associates (Bernthal, Green & Revzin, 1951) maintained that bradycardia resulted from chemoreceptor stimulation: Neil (1956)

showed that in cats there was certainly no carotid chemoreflex tachycardia, although he did not elicit bradycardia. There has since been ample confirmatory evidence that the primary effect of carotid body stimulation is vasoconstriction, with a secondary bradycardia accompanying the rise in blood pressure. Downing et al (1963) found the increased cardiac sympathetic activity in hypoxia (p.5/13) to increase still further if the sinus nerves were cut, indicating a chemoreflex tendency to decrease the heart rate: this confirmed their findings during perfusion of the carotid sinus (p.5/13). Similarly Scott (1959) and Daly & Scott (1962) using dogs, demonstrated the primary bradycardia effect, attributing tachycardia to a secondary effect of ventilation.

This evidence is concerned almost entirely with the carotid bodies. Comroe demonstrated in 1939 that the aortic chemoreceptors had proportionately greater cardiovascular than ventilatory effects, compared with the carotid bodies. He has more recently shown that in cats hypertension and tachycardia follow aortic chemoreceptor stimulation whilst hypertension and bradycardia follow carotid stimulation, by nicotine. The two effects were separated by interposing a delay coil. The result was the same during artificial ventilation. (Comroe & Mortimer, 1963), Daly & Ungar (1966) have clearly shown that the aortic and carotid chemoreceptors have similar effects in producing vasoconstriction and raising blood pressure, when the counteracting influence of greater ventilatory stimulation via carotid chemoreflexes is prevented. These authors do not, however, refer to heart rate changes, and it is not possible to see them on their published chart records. It could be that Comroe's contention remains unanswered, and that tachycardia could still be attributed to the aortic chemoreceptors.

b) Pressure receptors

Increased pressure in the right heart has been mentioned as a possible cause of tachycardia.

Increased pressure in the left atrium could possibly follow from pulmonary vascular changes, and could excite left atrial receptors as described by Ledsome & Linden, 1967, leading to increased heart rate. However, it is stated that increased left atrial pressure does not occur in hypoxia (Aviado, 1960).

c) Vagal afferents from the lungs

The increase of heart rate attributable to increased ventilation in anaesthised hypoxic dogs, is prevented by denervation of the lungs (Scott, 1959). In cats, Downing et al (1963) found no change in the increased cardiac sympathetic nerve activity in hypoxia when the vagi were cut, and concluded that it did not depend on vagal afferents.

In man there is some direct evidence that neither carotid chemoreflexes nor ventilatory increase are implicated in the production of tachycardia: two patients whose carotid bodies had been surgically removed showed abolition of the ventilatory response but retention of the heart rate response in hypoxia (Holton & Wood, 1965).

In summary, published reports suggest that the tachycardia of hypoxia is probably;

1. mediated by the sympathetic (though decreased vagal tone is not excluded)
2. due to a direct action of low PO_2 in the medulla or brain stem (and/or spinal cord) with possible contributions from:
3. a circulating cardiotonic agent, released by the adrenals
4. a link with increased ventilation

The possibilities appear to remain open that there could be;

5. a direct effect on the SA node
6. an aortic chemoreflex
7. an effect on heart rate secondary to the change in pulmonary haemodyanics.

Methods of study applicable to man

1. Timing of responses

This approach has already been suggested as appropriate to a situation where it is not possible to study component parts of an overall response by anatomical separation.

The methods of Dejours and his colleagues, as applied to the timing of ventilatory responses, have been reviewed. In brief, it is possible in man to recognise a response to arterial chemoreceptor stimulation by means of breath-by-breath measurements immediately after a change in inspired gas.

Proposed studies

By recording heart rate as well as ventilation during hypoxia, and during an abrupt change to hyperoxia, it seemed likely that supportive evidence might be found for or against certain sites of action. It should be possible also to investigate

i) any cause-effect link with ventilation: if the heart rate changes were to any extent secondary to ventilatory changes, then they must accompany or follow the ventilatory changes.

ii) primary chemoreflex effect: an abrupt change in oxygen tension might transiently reveal the chemoreceptor effect on heart rate (if the overshadowing opposite effect were NOT linked to ventilation, and if the site of action for inducing it took longer for the blood to reach).

Since planning and carrying out the experiments to be described, it was brought to my notice that Dejours et al (1958) did in fact include heart rate in their observations; although it was not their primary concern, they suggested that this might be a useful way of investigating heart rate response. When a brief account of the present experiments was published (Jennett, 1969), Dejours told me of almost exactly similar experiments carried out by Hernandez-Martinez (1967).

2. Study of patients with pathological or surgical destruction of some part of the pathways concerned.

ABBREVIATIONS USED IN SECTION 5

Heart rate

h	frequency of heart beat, beats per min
h_I	h at peak of inspiratory transient
h_E	lowest h in expiration

(see p. 5/45)

Time periods

A H O: Air, Hypoxia, Oxygen: whilst breathing air, 10% oxygen, 100% oxygen

Ref. Last two min breathing air, before test

$H_{2,3}$	Second + third	min of hypoxia
$H_{\sqcap}$	Last	min of hypoxia

$O_{\frac{1}{2}}$	First half-minute	after first inspiration of 100% oxygen
O_1	Second half-minute	after first inspiration of 100% oxygen
O_{10+}	From tenth to sixtieth sec	after first inspiration of 100% oxygen

(see p. 5/48)

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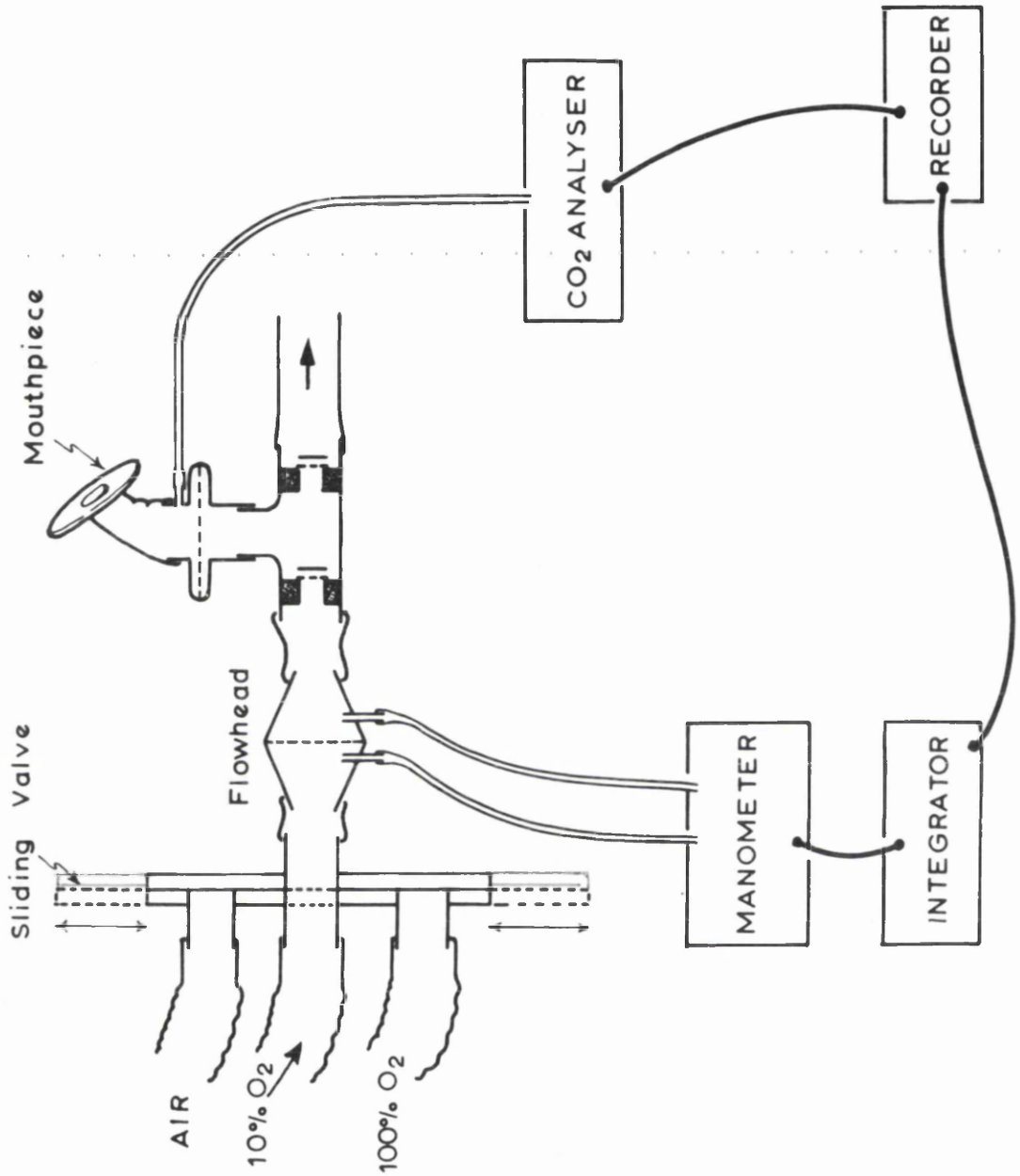
Fig 5:1



Arrangement of apparatus for hypoxia - re-oxygenation experiments

EXPERIMENTS ON THE RELATIVE TIMING AND EXTENT OF VENTILATION AND HEART RATE CHANGES DURING HYPOXIA AND REOXYGENATION

METHODS

The several components of the apparatus for respiratory monitoring and for changing the composition of the inspired gas were as described in a previous section and the particular arrangement for these experiments is shown opposite (Fig.5.1). In addition, heart rate was recorded continuously, beat by beat, and an additional ventilatory parameter was used.(p 5/25)

Inspired gases

Douglas bags were filled well in advance of experiments so that gases were at room temperature. Hypoxic mixtures for all experiments were 9.5 - 10% oxygen in nitrogen, made up on a manifold, analysed by Lloyd-Haldane apparatus, and the analysis checked with the Servomex analyser before use. This mixture, air, and 100% oxygen respectively were connected to the appropriate inlets of the sliding valve.

Heart rate recording

ECG electrodes. In earlier experiments these were conventional metal electrodes applied to the two

wrists and one ankle; in later experiments miniature gold-plated cup electrodes (Grass) were used on the chest (positions 2 and 4, and reference electrode on the back).

ECG was displayed on a Tektronix 502 C.R.O., which was connected to the Devices Ratemeter, of range 8-100 and 20-500/min. This instrument gives an output proportional to the inverse of the interval between each beat and the preceding one: in this instance the R wave is used as the trigger. The ratemeter was calibrated with a Devices pulse generator. The heart rate was recorded directly on one channel of the U-V recorder.

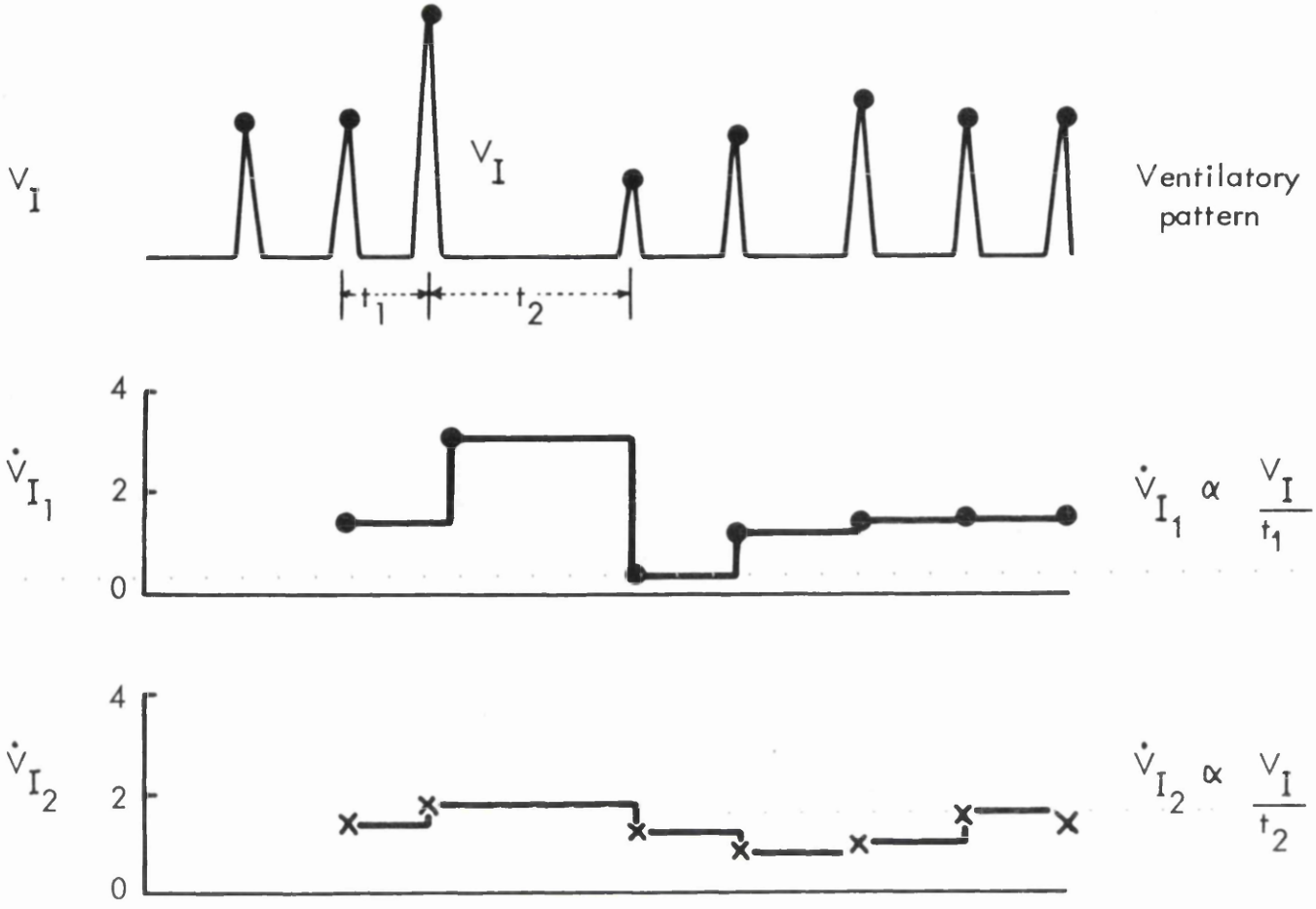
Ventilatory recording

Inspiratory pneumotachography was employed, as described.

Breath-by-breath ventilation computation

In earlier experiments a tidal trace only was used (e.g. Fig. 5:5) but later direct breath-by-breath ventilation computation became available, developed at my suggestion by Dr. J.R. Greer, as an additional facility associated with his own manometer and integrator, and

Fig 5:2



Computation of breath-by-breath ventilation:

Top: Hypothetical sequence of breaths; successive inspired tidal volumes recorded.

t_1 : interval between breaths, preceding breath of volume V_I

t_2 : interval between breaths, following breath of volume V_I

Middle: Output which would result from computing breath-by-breath ventilation as proportional to $V_I \div t_1$

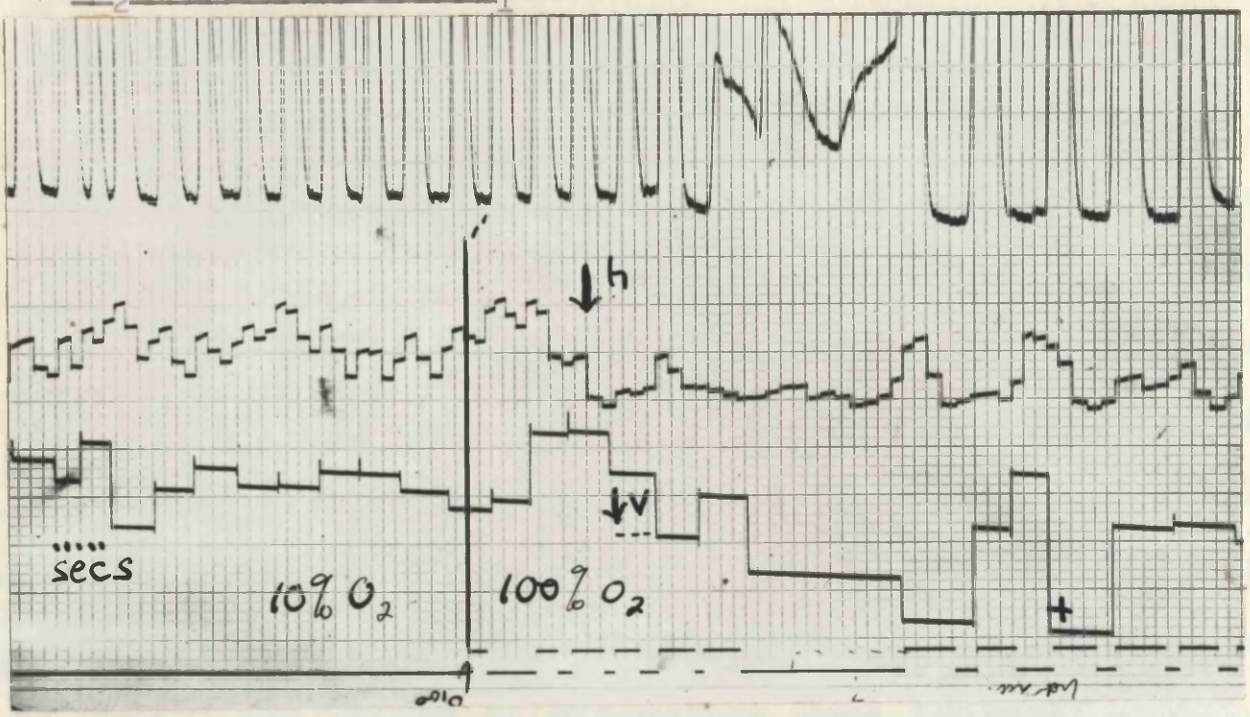
Bottom: Output which results from the present method of computing breath-by-breath ventilation as proportional to $V_I \div t_2$

specifically for use in experiments of this nature (Greer & Jennett, 1969 appended). The meaning of this can be described as follows: at the end of each inspiration the integrated tidal volume (V_T) is stored; at the end of the next inspiration, an output is given proportional to the preceding V_T divided by the time interval between the two breaths (V_T/t). Thus, for each breath this has the dimensions of "minute volume" - volume per unit time, or tidal volume×frequency. It will be abbreviated as $\dot{V}_I$.

It might seem preferable to relate the volume of each breath to the interval which precedes it (t_1) rather than to that which follows it (t_2), if respiratory centre activity leads to a breath of a given size, starting at a given time after the previous one. If on the other hand a deep breath <u>followed</u> by a long interval is represented by an output which compounds these two, this really assesses two successive states of ventilation: a deep breath due to some stimulus, and a compensatory pause associated with the deepness of the breath itself. But this latter V_T/t_2 measurement can be held to be a useful indication of activity leading to just such stable compensation: a V_T/t_1 output would vary much more breath by breath, when frequency and tidal volume vary, whereas,

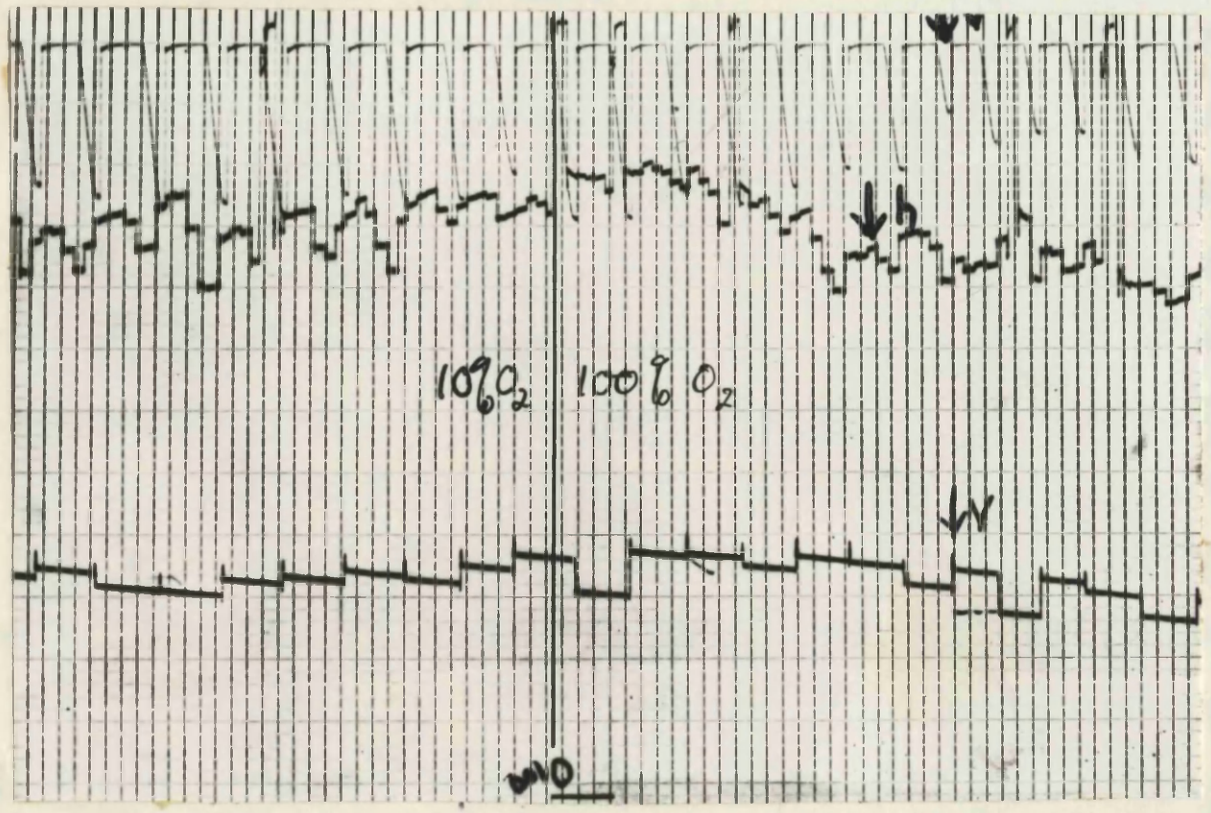
Fig 5:3 Examples of traces , using breath-by-breath ventilation output. Two records of the change from 10% to 100% O_2.

a) CO_2, heart rate, and $\dot{V}_I$



Absence of end-tidal plateau 6th-7th breath after 100% O_2 confirms reduction in tidal volume, but there is a discrepancy in the $\dot{V}_I$ trace at +

b) Tidal volume, heart rate, $\dot{V}_I$.



Each new deflection on $\dot{V}_I$ trace can be seen to correspond with the end of an inspiration.

In both these traces, heart rate can be seen to decrease before $\dot{V}$.

if the ventilation-rate is being kept steady over a few breaths, then the V_T/t_2 output will be relatively steady. Fig.5:2 shows the different outputs which would be recorded with different changes in pattern, using the two computations respectively. V_T/t_2 was electronically expedient, and on the above grounds, physiologically defensible as a guide to ventilatory activity. The first sign of response to a particular stimulus can be seen separately on the concurrent tidal trace. For some experiments this $\dot{V}_I$ was the only ventilatory measurement used (e.g. Fig.5:3a). At that time there were only three channels available for recording and the other two were used to record end-tidal CO_2 and heart rate. However, it became evident that a simultaneous tidal trace was necessary in order to distinguish and accurately time changes of interval and changes of tidal volume; also the presence of both traces enabled identification of artefacts due to swallowing, resetting of the integrator unit, and so forth (Fig.5:11 p.5/45) Until a fourth channel became available, some experiments were carried out without continuous CO_2 measurements (Fig. 5:3b), but with occasional spot-checks; this accounts for the incompleteness of some data.

CO_2 recording

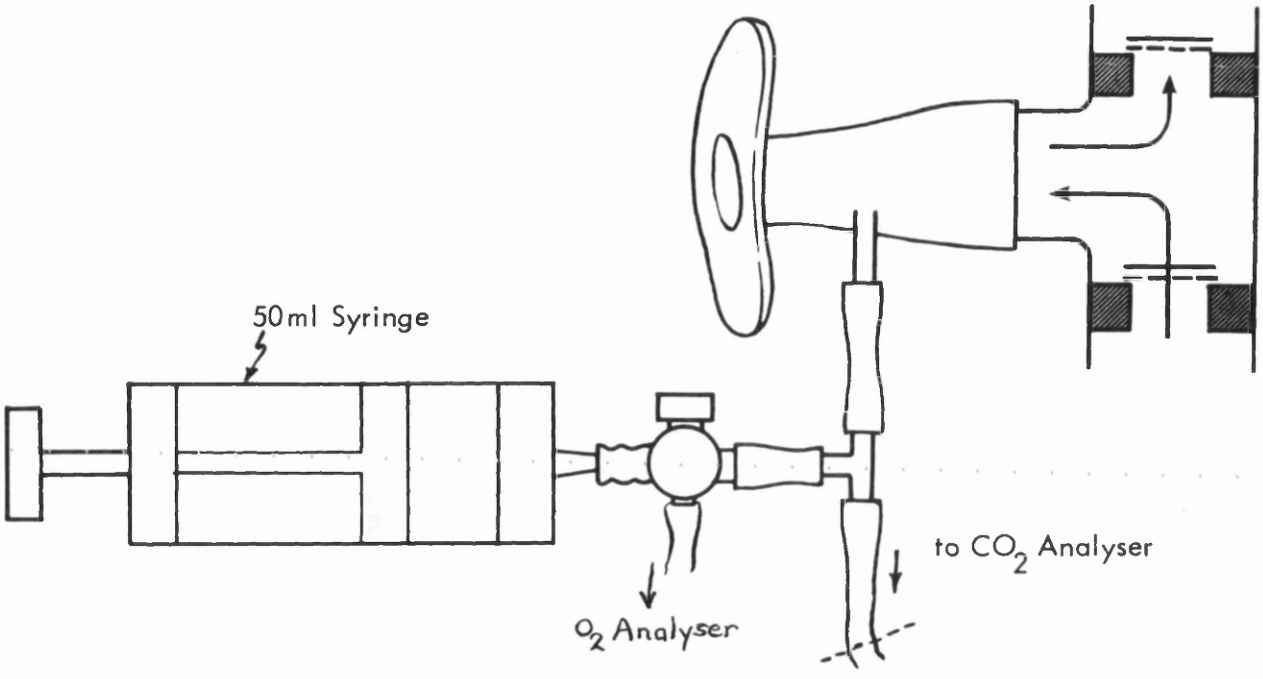
Sampling and analysis were carried out as described (p.2/7) with the URAS analyser. In the later series, CO_2 was sampled at the mouth rather than beyond the expiratory valve.

Measurement of alveolar oxygen tension

Ideally, end-tidal PO_2 should be monitored in these experiments. The apparatus for this was not available, and indeed presents considerable problems unless expense is no object. The possibility of modifying the Servomex to read out continuously was discussed with B.B. Lloyd, since this had been done in the Oxford laboratories, but it appeared that this would not justify the technical time and effort involved. Ear oximetry would have been an approximate guide to changes in oxygenation, but not accurate enough to make precise statements about the levels attained. This facility was deferred until a relatively reasonably priced rapid oxygen analyser, fuel-cell or paramagnetic type, becomes available. In any case, the chief aim of these experiments was not essentially to define what happens quantitatively to ventilation, or to heart rate, at a particular level of hypoxia, but to discover

Fig 5:4

ARRANGEMENT FOR END-TIDAL SAMPLING FOR P_{A-O_2}



Gas was withdrawn during the latter half of expiration over 3-4 breaths, with the CO_2 sampling flow interrupted. Continuous sampling for CO_2 was then resumed whilst the syringe was emptied into the oxygen analyser.

the relationship between the changes in each to those in the other, and the timing of the changes in each. It was therefore felt not to be a serious handicap not to have PO_2 measurements in each and every experiment. Instead, in several individual representative experiments, careful end-tidal samples were taken at different times and analysed.

Method

A 50 ml syringe was connected by three-way tap to the expired air sampling tube leading from the mouthpiece to the URAS analyser (Fig.5:4). The CO_2 sampling flow was switched off, and the syringe was filled by withdrawing during the second half of expiration over the period of 3-4 breaths - by hand, and by watching the subject's breathing pattern. During inspiration and the first half of expiration, the piston was very slightly advanced to ensure no intake. When the syringe was full, CO_2 monitoring was restored, and the sample was put through the Servomex; by preliminary tests it had been found that this volume was sufficient for a steady reading if the apparatus dead space was cut down as much as possible and if the sampling cell was already flushed through with gas of composition near to that of the sample. Accordingly a

7-10% oxygen mixture was put through initially, and the inlet closed off between analyses to avoid intake of air.

Blood pressure

No facility was available for the monitoring of blood pressure, and occasional sphygmomanometer readings would have been likely to cause variations in heart rate by disturbing the subject. Such readings were therefore deliberately not undertaken. There is good evidence from the literature that the inhalation of 10% oxygen does not cause any change in blood pressure in man (e.g. Richardson et al. 1966, 1967); however, these measurements had not been made with a view to temporally relating any small changes to changes in heart rate.

On a few separate occasions the subjects who were most accustomed to the apparatus were studied again with the same regime of gas changes, and sphygmomanometer readings taken as frequently as possible during control, hypoxic, and re-oxygenation periods. There were no significant changes, but this method is admittedly relatively crude, both in timing and in accuracy, compared with recording the heart rate and ventilation and it is not taken as demonstrated that no definite, if transient change occurs. The results are shown in Fig. 5:29, p.5/84 and the possible significance of changes more fully discussed.

THE TWO SERIES OF EXPERIMENTS

The experiments fall into two groups: in the first series observations were made during hypoxia of varying duration. On the basis of these a particular routine was selected, and later applied more extensively and further developed in the second series.

Experimental routine common to both series

The subjects were all normal healthy men. They were studied in a resting, but not basal state, at least two hours after the previous meal. They lay on an examination couch, or, later, in a reclining chair with the legs near horizontal and with the head raised.

FIRST SERIES

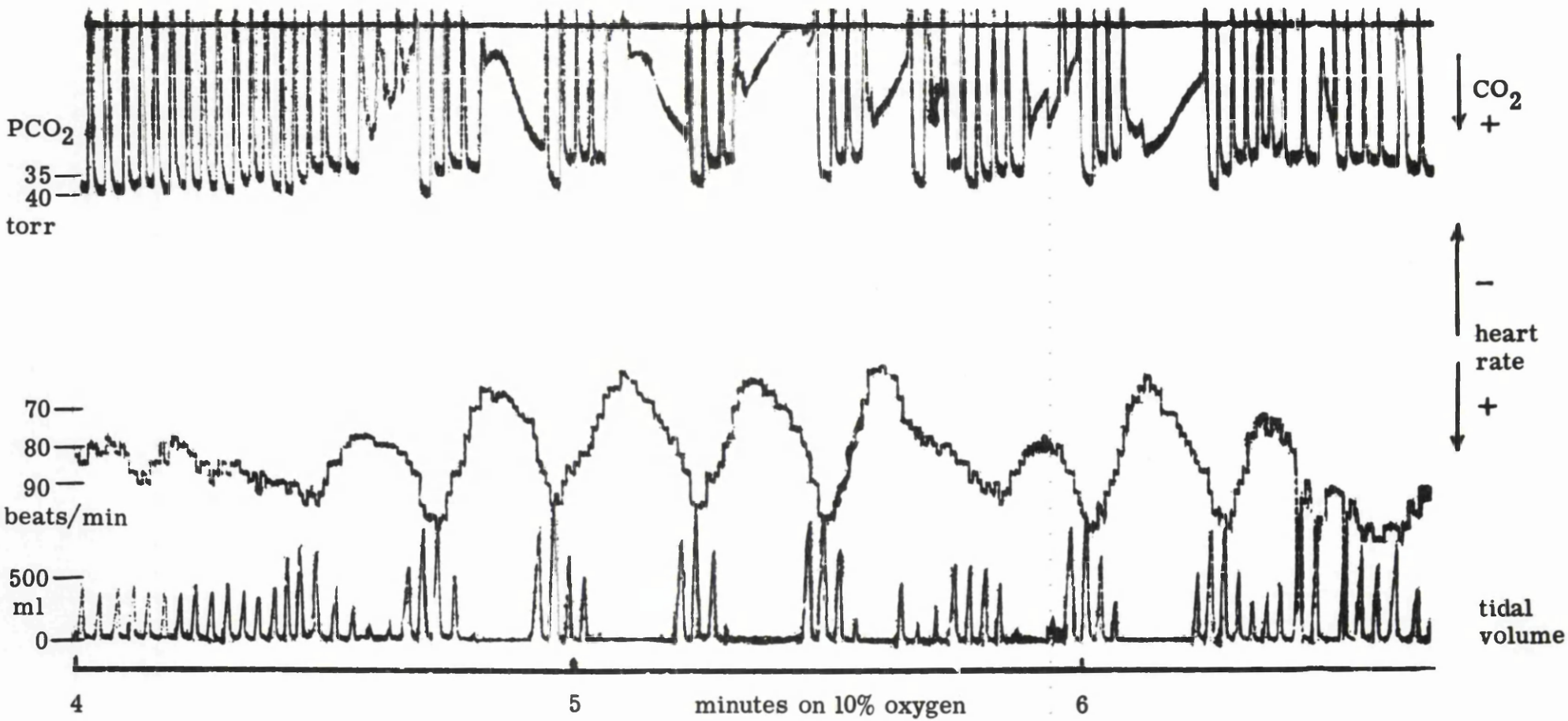
Subjects and methods

Three subjects (W, C, K) were studied repeatedly. All were healthy young men, and used to the equipment. One of the three was in athletic training, (K).

Several sequences of changes were carried out, some combined with observations on the effects of 100% oxygen, reported elsewhere. The experiments which form the group to be described followed this routine:

1) The subject breathed room air until reasonably

Fig 5:5



Subject C: Periodicity in heart rate and ventilation developing after 4 minutes hypoxia.

steady levels of heart rate and ventilation were obtained: air from a bag was then inspired for 2-3 minutes.

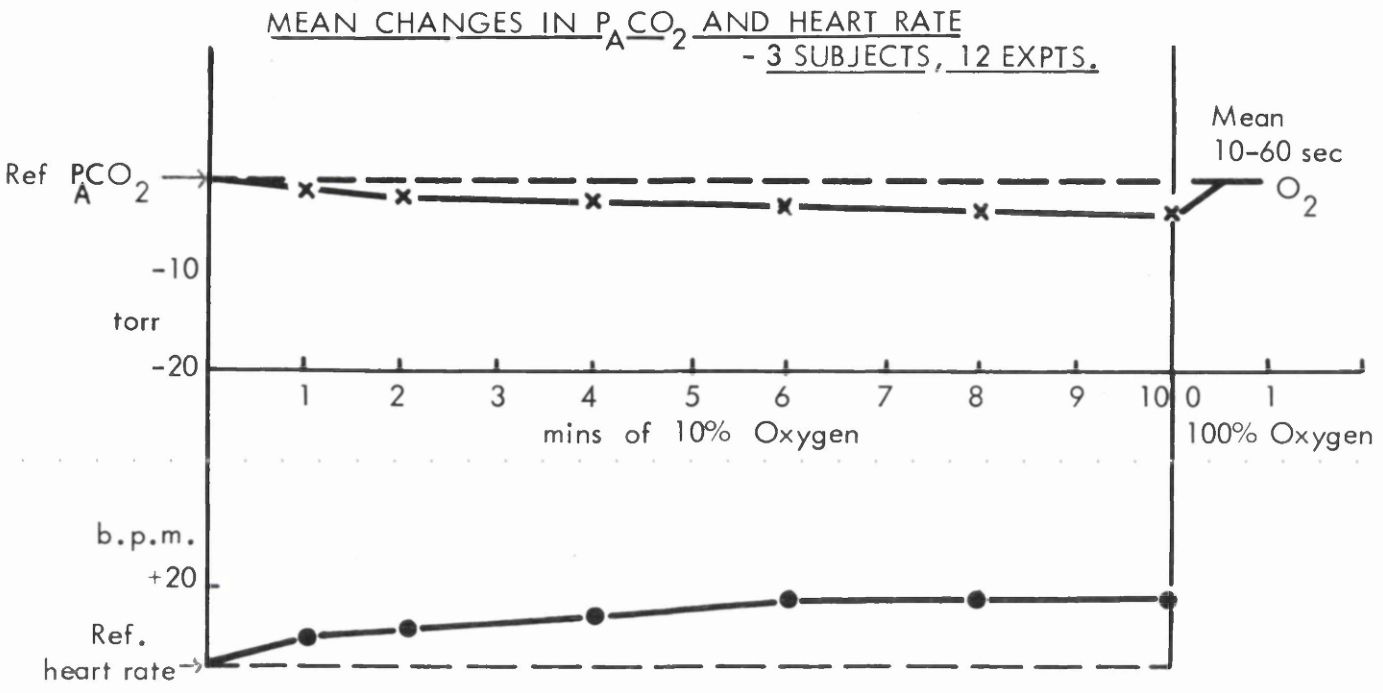
2) A switch was made during an expiration to 10% oxygen in nitrogen, and this was inspired for 10-13 minutes.

3) A switch was made to 100% oxygen, or in some instances to air. From these studies a total of 12 complete and comparable sequences were available for analysis - 4 on each subject using oxygen for reoxygenation, and 5 more using air.

Results of the First Series

In two of the subjects (W, C) the pattern of breathing usually became irregular after 3-4 minutes breathing 10% oxygen. In C particularly, this irregularity several times took the form of typical periodic breathing. (Fig. 5:5). The behaviour of heart rate during this pattern was of interest in itself, but the irregularity of both heart rate and ventilation complicated any analysis of the change on abrupt re-oxygenation. A mean level could be found for, say, the final minute of hypoxia, and compared with the mean level during the first minute of re-oxygenation, but it

Fig 5:6


MEAN CHANGES IN P_ACO_2 AND HEART RATE
- 3 SUBJECTS, 12 EXPTS.
Mean
10-60 sec
O_2
Ref P_ACO_2
-10
torr
-20
1
2
3
4
5
6
7
8
9
10
0
1
mins of 10% Oxygen
100% Oxygen
b.p.m.
+20
Ref.
heart rate

was not possible clearly to define the timing of the changes, because the change concerned was of a smaller order than the swings during periodicity. (Fig.5:7b,overleaf).

The third subject (K) did not show this irregularity.

1) Changes during hypoxia

On average, and in each subject, ventilation and heart rate increased (Fig.5:9).

A detailed method of analysis, involving measurement of the size and duration of each breath, and the mean (tidal volume xf) for each three breaths together with the mean heart rate over three breaths, was applied to a few runs. (The breath-by-breath ventilation rate computer was not yet available for this series). The value of this procedure was doubtful in relation to the time it took: there were no measurements of alveolar oxygen concentration, and both the time course and the degree of the hypoxia were therefore unknown variables between subjects and occasions, This was not therefore pursued.

The increase in heart rate tended to continue longer than the increase in ventilation, which was usually maximal within the first three minutes. Fig.5:6 shows mean change in heart rate and change in end-tidal PCO_2 plotted against time, and it appears that on average the two changes were approximately concurrent. End-tidal PCO_2 was used here as the index of change in alveolar ventilation, (but may not be an accurate guide - p.5/38

Fig 5:7a

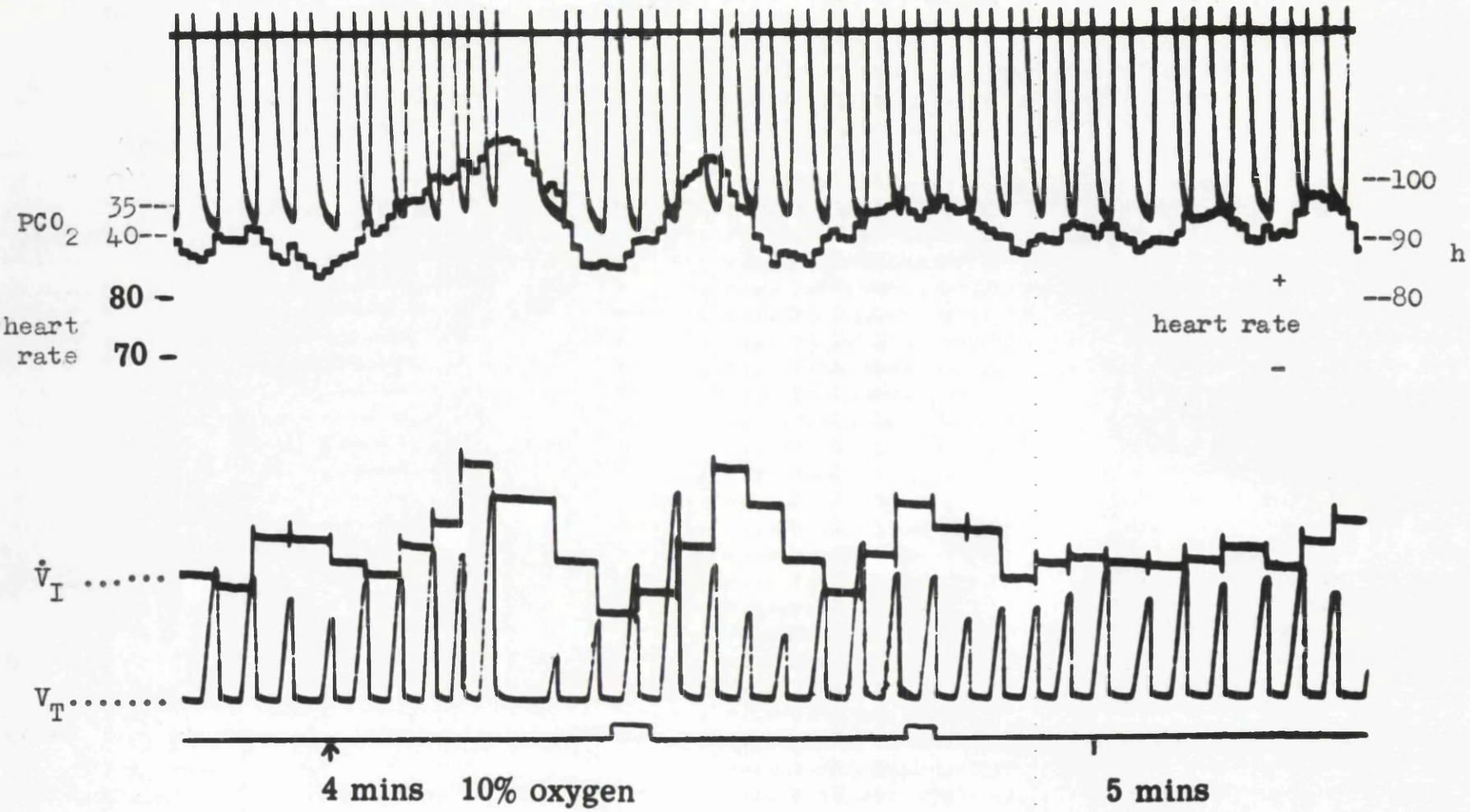
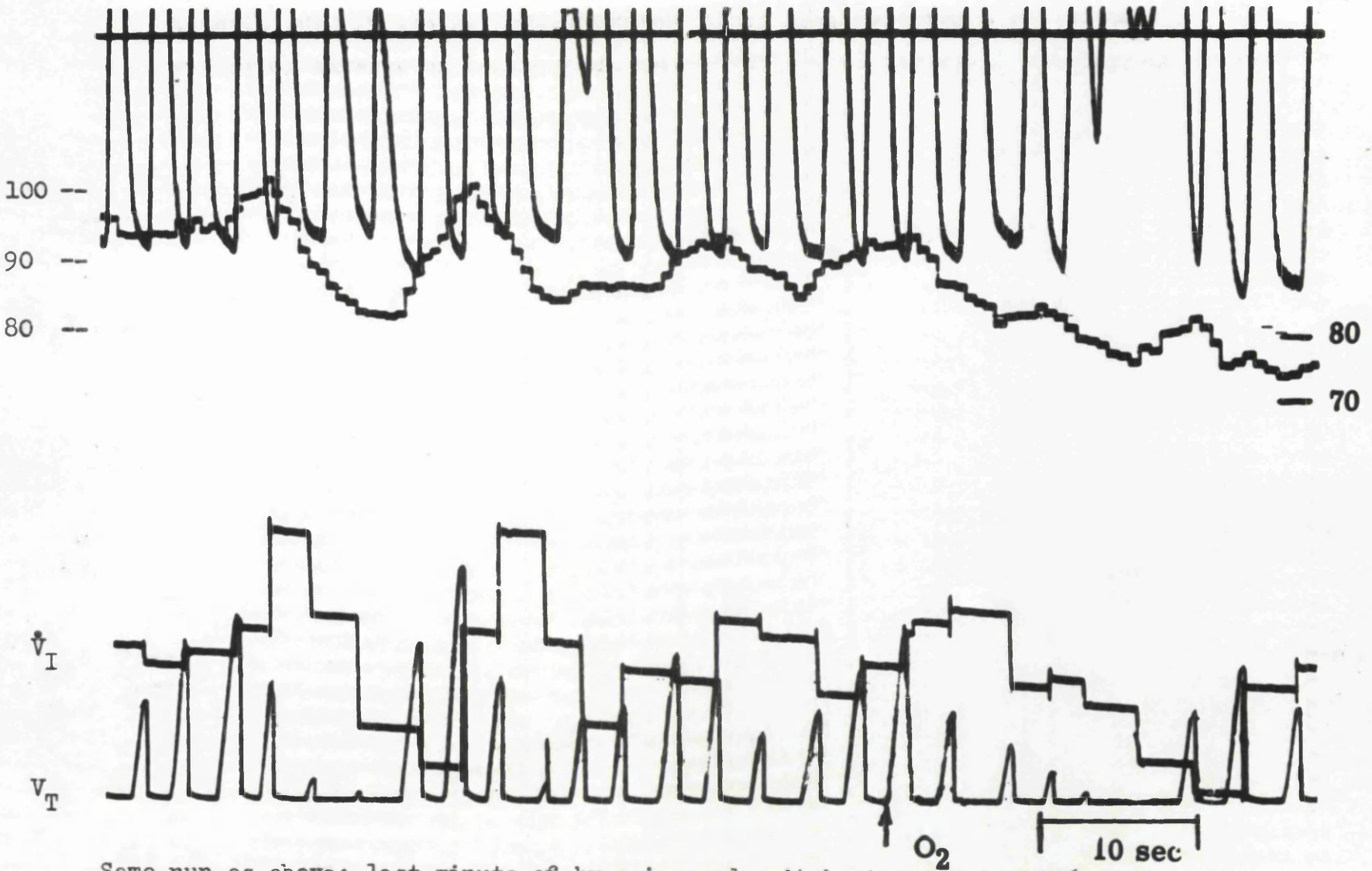


Fig 5:7a

A further example of periodicity developing during hypoxia ; the pattern is less regular than in Fig 5: but continues until reoxygenation - see below.

Fig 5:7b



Same run as above: last minute of hypoxia, and switch at arrow to 100% oxygen. Concurrent decrease in heart rate and ventilation within 15 seconds.

Fig 5:7b

2) Changes during reoxygenation

It was decided that attention was best given to the detailed study of this change. Unlike the induction of hypoxia, the timing, if not precisely the extent, of the step-change to hyperoxia was controllable and relatively comparable between different subjects and occasions. It was calculable that the hypoxic stimulus at the chemoreceptors would be cancelled out completely and abruptly within the first three breaths of oxygen (see p. 5/72).

Tidal volume and frequency usually decreased within the first few breaths as in the "oxygen test" of Dejours et al (1958); this was reflected in a small increase in end-tidal PCO_2 in breaths which were large enough to reach an alveolar plateau (Fig. 5:3a, 5:7b). Heart rate also decreased, usually more or less at the same time as ventilation, within the first 20 sec (Fig.5:7). Mean changes are shown in Fig. 5:9.

The most striking observation, however, in this series, was that in subject K, in whom the changes could be seen clearly on account of the absence of irregularity during hypoxia, a step change in heart rate was consistently seen unrelated to, and before, a change in tidal volume.

Fig 5:8

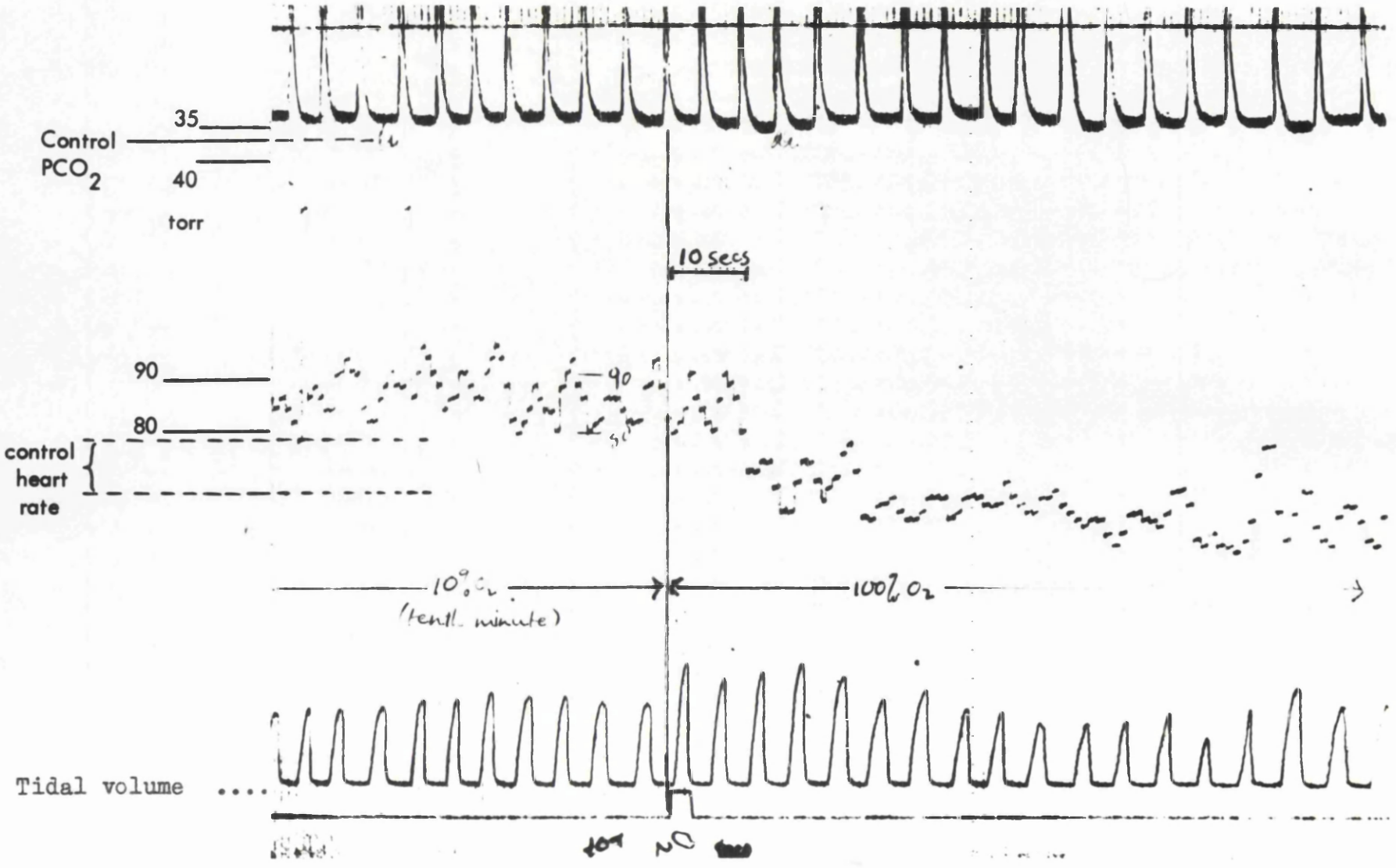


Fig 5:8

Subject K: The left third of the trace is from the tenth minute of hypoxia (breathing 10% oxygen) Vertical line represents switch to 100% oxygen. There is an abrupt decrease in heart rate, dissociated from any ventilatory change.

The clearest example of this is shown in Fig.5:8 (opposite) where the fall in heart rate is seen to occur 12 secs after the beginning of the first inspiration of oxygen. For some reason, probably unrelated to blood gas levels, several deeper breaths happened to be taken just at this time, and the change in heart rate can certainly not be associated with a ventilatory change. Tidal volume is seen to decrease after the first 25 secs and there is a further small decrease in heart rate with a reduction in the respiratory swing. Although this was the most convincing example, a change in heart rate after a comparable interval occurred in each experiment on this subject, whereas the changes in tidal volume tended to be minimal or delayed (Fig.5:10)

Detailed analysis of results

All values used for the diagrams (Fig.5:9, 10) were derived as follows:

1. Ventilation - $\dot{V}_I$ - was calculated as the measured cumulative tidal volume per minute for the appropriate period, in arbitrary units (mm deflection per min). (In some studies expired volume was also measured, and an approximate calibration factor for

VENTILATORY AND HEART RATE CHANGES DURING HYPOXIA AND REOXYGENATION

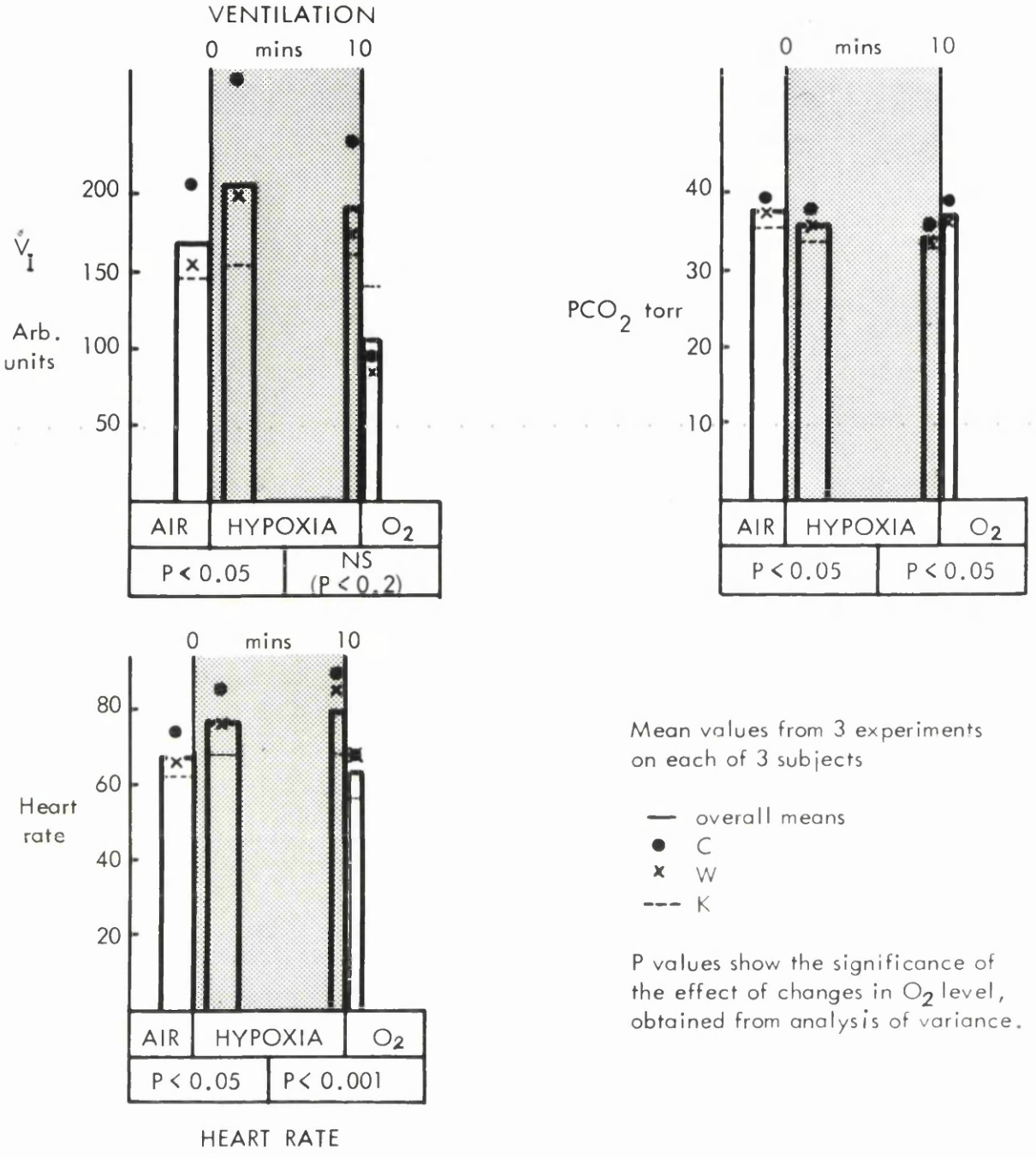


Fig 5:9

Shaded area represents period breathing 10% oxygen.

Width of columns represents period over which mean measurements were taken: 2 min on air; 2nd and 3rd min on 10% oxygen; last min on 10% oxygen; 10th-60th sec after change to 100% oxygen.

$\dot{V}_I$ derived. This gives a mean overall resting ventilation of 8.4 L/min).

2. P_ACO_2 — A mean value for the selected periods was found by taking the mean of each end-tidal value, ignoring breaths where an alveolar plateau was not reached. % values were converted to torr with reference to the ambient pressure.

3. Heart rate — The actual number of beats per minute was counted from the beat-by-beat heart rate trace.

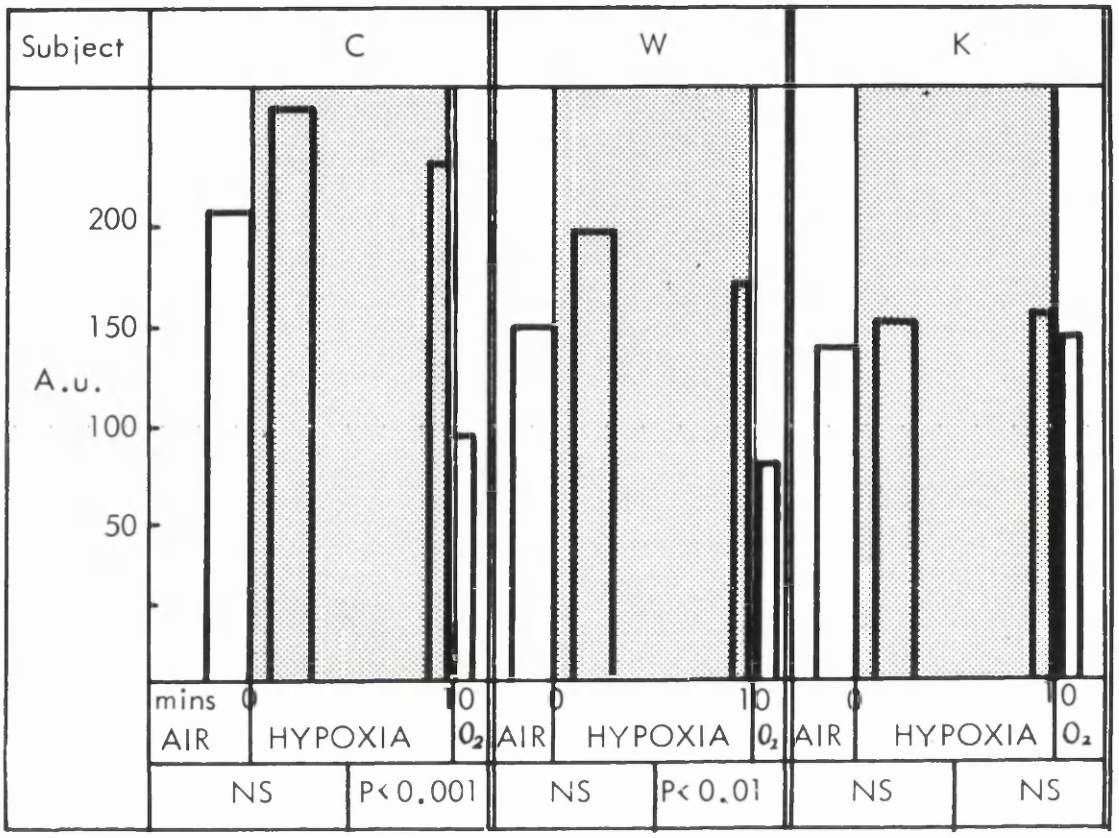
Analysis of variance

3 experiments on 3 subjects were used for this. (3 was the maximum common number of complete experiments). Where more than 3 complete experiments were available for one subject, 3 were selected randomly.

The means for all subjects, and for each separately, for each period are shown in the diagrams (Fig. 5:9). The "AIR" value represents the last 2 min before starting to breathe 10% oxygen; the earlier hypoxic value the second and third minutes on 10% oxygen; the later hypoxic value, the last minute of 10% oxygen; the O_2 value, the period

VENTILATORY CHANGES DURING HYPOXIA AND REOXYGENATION

Mean values from all experiments on each subject separately



P values show significance of changes obtained from Student's t-test for difference between means

Fig 5:10

Measurement periods as in 5:9.

A.u. : arbitrary units for ventilation.

from 10 to 60 seconds after the beginning of the first breath of oxygen. (These standard periods were selected in the course of analysing the later series, p 5/48 and applied retrospectively to this series, so that the results would be comparable).

The effects of change in O_2 level were analysed separately for air ⟶ hypoxia (2nd - 3rd min) and for hypoxia (last min) ⟶ O_2. The other variable was the difference between subjects, and the three repetitions on each subject were taken as replications.

Ventilation: Air ⟶ hypoxia: The effect of O_2 level was significant, as also was the variability between subjects.

Hypoxia ⟶ O_2: The effect of O_2 was non-significant - obscured by the interaction with subjects: i.e. different subjects showed different changes during reoxygenation.

Because of this obscuring effect, all available experiments were used to obtain mean values for each subject separately (Fig 5:10). This showed a

significant effect of reoxygenation on ventilation in two of the subjects, but not in the third. Within subjects, the effect of the change from air to hypoxia was too variable to show significance in the small number of experiments.

$\underline{PCO_2}$: Air ⟶ hypoxia) The effect of O_2 level was
Hypoxia ⟶ O_2) significant, and did not vary between subjects, although the actual PCO_2 level varied between subjects.

Fig. 5:9shows that there is an apparent discrepancy between the direction of changes in PCO_2 and ventilation from the beginning to the end of hypoxia. On average, and for each individual, the PCO_2 apparently continues to fall although the measured ventilation also falls on average, and in 2 of the 3 subjects. The likely explanation is this: the two subjects who show the discrepancy are those who were breathing irregularly in the late hypoxic period; this means that several tidal volumes were too small to give an alveolar reading for CO_2, and during these PCO_2 would in fact be at its highest; also the uneven pattern of breathing makes the mean of breath-by-breath end-tidal values less valid as a guide to arterial PCO_2, or indeed of mean alveolar PCO_2 over a given period. Both these factors would make it likely that the PCO_2 would be underestimated. It seems more likely, therefore, that the measured decline of ventilation towards the end of the ten minutes is real and the estimated decrease in PCO_2 is not.

Heart rate: Air ⟶ hypoxia: The effect of O_2 level was significant.

Hypoxia ⟶ O_2: The effect of O_2 level on heart rate at reoxygenation was the most significant in the whole analysis.

There was no statistically significant change in any parameter between the earlier and the later hypoxic period: mean of all changes in heart rate showed that 65% of the total 10-minute increase had occurred by the third minute.

Timing of responses to reoxygenation

For ventilation: In two of the subjects it was not possible clearly to define the onset of depression at reoxygenation, because of the very great variability of the tidal volume and frequency in the late hypoxic period.

For subject K: A clear fall was definable at a mean period of 28 seconds after the first breath of oxygen, with a range of 22-37 seconds. This subject, however, showed the least dramatic changes in ventilation (Fig. 5:10).

For heart rate: It was possible to define a fall in heart rate more clearly than a fall in ventilation, even for the two subjects with periodic breathing. The mean time for all three subjects was 16 sec (range 10-24) to the first clear reduction after the beginning of the first breath of oxygen. For subject K alone the values were 14, range 10-21 sec.

In subject K the reduction in heart rate was always clearly defined, and always preceded any reduction in ventilation, by a mean of 14 sec.

Conclusions from these studies

It appeared likely that further modified experiments would give useful information if particular attention were given to the timing of changes after the switch to oxygen.

If heart rate could be shown consistently to fall with a time lag similar to that for the change in ventilation, this could support either a similar reflex mechanism, or one for heart rate which was of similar timing to the chemoreflex effect on ventilation but with different receptors; or a direct effect of sudden oxygenation on the lungs or heart taking a similar time to act. It would be evidence against a site of action with a longer delay, which might include the medullary centres.

If the change in heart rate could be shown to be commonly dissociated from the change in ventilation, although on average of similar timing, this would add to existing evidence against any direct link with ventilatory change.

SECOND SERIES OF EXPERIMENTS

Introduction

The first series had shown that a significant increase in heart rate, and commonly also in ventilation, had already occurred within the first 4 minutes of inhaling 10% oxygen, and that irregularity was liable to develop thereafter. In this later series, therefore, it was decided to expose the subjects to the hypoxic mixture for only 3-4 minutes, and then abruptly reoxygenate with 100% oxygen. A true steady state was unlikely to be attained at 3 minutes, but there was reason to believe that the change in P_AO_2 would have reached 80-90% of its ultimate value (Opitz, 1941); also it was known from the earlier series that most of the changes evident at 10 minutes had already occurred at 3 minutes. In any case it can be argued that when responses to an abrupt change are being studied, the observations of most interest are those which are made during the first disturbance of the status quo, before secondary effects have complicated the picture (Dejours, 1957). Even were the level of hypoxia, and the heart rate and the ventilation still changing at 3-4 min, the chief interest lay in the timing of the

PLAN OF AN EXPERIMENT

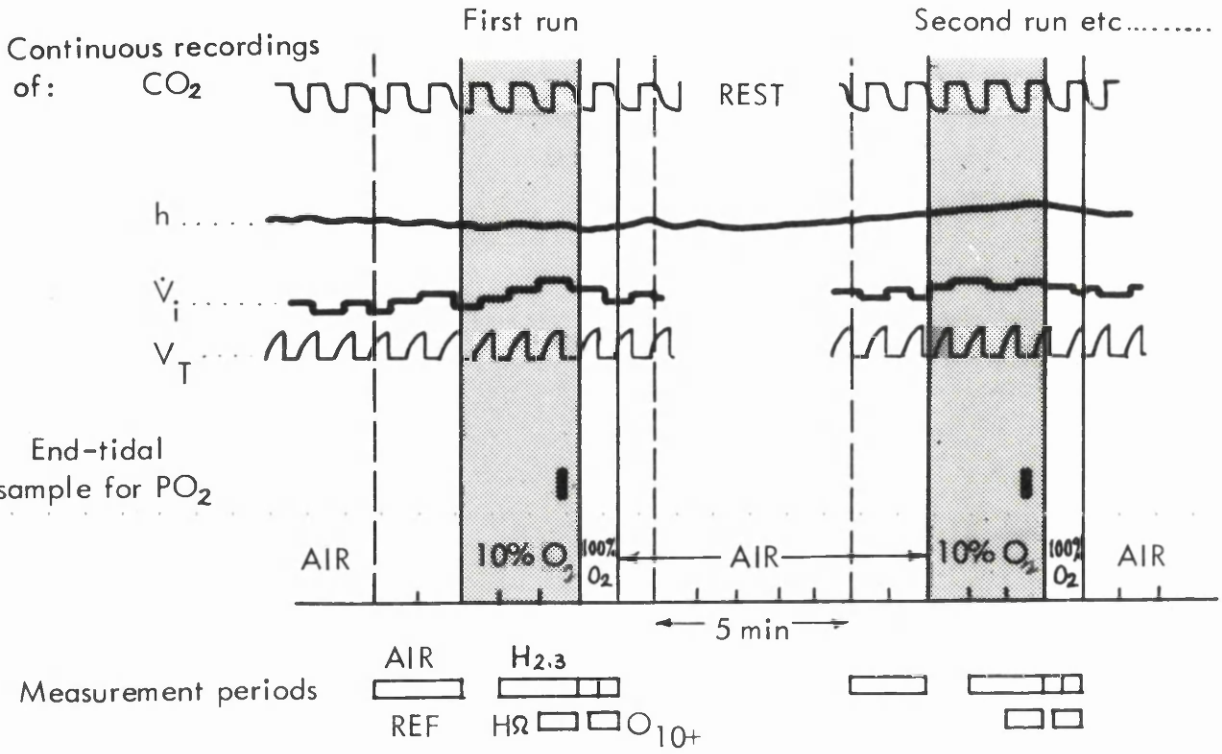


Fig 5:11

responses of each respectively to reoxygenation; the steadiness of the hypoxic state was theoretically not relevant to this, provided that a sufficient increase had been obtained to show statistical significance compared with control values and provided that such increase was stable enough over the period of the final minute of hypoxia to show significantly the decreases on reoxygenation. The early series had shown that these conditions were likely to be satisfied.

Methods and subjects

The plan of a single run (Fig.5.1) was therefore:

After settling to steady levels the subject breathed

1) Air from a bag for 2 minutes
2) 10% oxygen in nitrogen for 3-4 minutes
3) 100% oxygen for 2, 1 or ½ minute. (These times became progressively less with successive sessions as it was realised that (a) no further information was gained, since reoxygenation was complete by ½ minute and (b) a shorter interval could be allowed between successive hypoxic runs if the oxygen period were kept to a minimum)

A series of sham experiments was carried out on 3 subjects: the procedure was exactly similar, but all bags contained air.

Table 5:1

Subjects and experiments in main (second) series

To show number of experiments on each subjects, system of numbering, and the experimental runs which were included in the different analyses of results.

Subjects : W, M , B, S, H.

First session, or occasion, and successive runs numbered thus:

1:1 , 1:2 , 1:3 etc.

Bracketed runs were excluded for various reasons from the overall analysis of mean results. (see text)

SUBJECT	W	M	B	S	H	
	W1:1	M 1:1	B 1:1	S 1:1	H(1:1)	first occasion
	1:2	1:2	1:2	1:2	(1:2)	analysis, p5/57
		(1:3	(1:3)	1:3	(1:3)	
			B(2:1)	S 2:1	H 2:1	second occasion
			(2:2)	2:2	2:2	3 occasions combined
					2:3	
			B(3:1)	S 3:1	H 3:1	third occasion
			3:2	3:2	3:2	
			3:3	3:3	3:3	
	5-run analysis p5/58		3:4			
			3:5			
				S 4:1	H 4:1	
					4:2	
					4:3	
					H 5:1	
					5:2	
Total runs	2	2(1)	6(4)	9	11(3)	30(8)

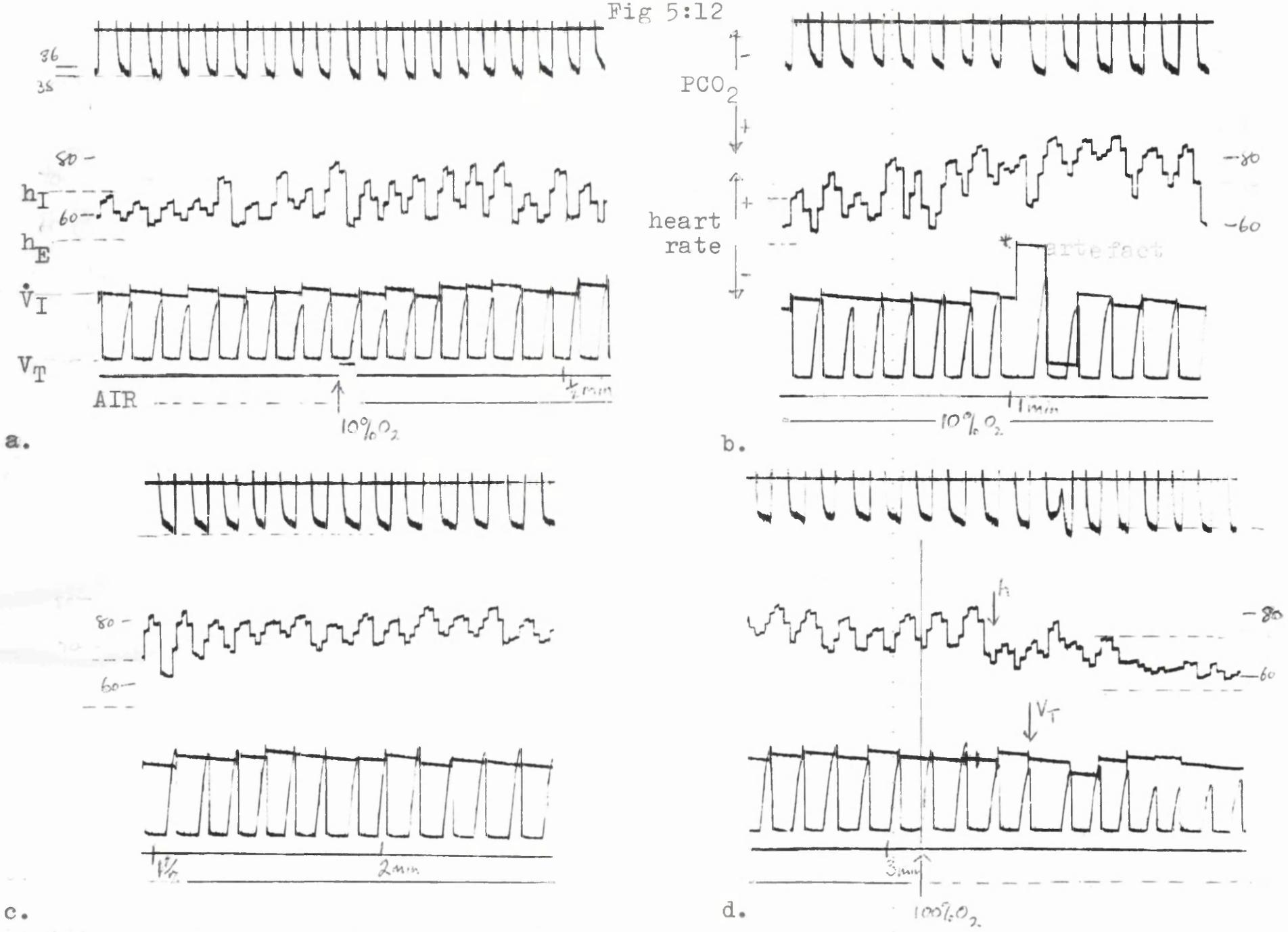
In experiments when alveolar oxygen was measured, samples were taken between 2½ and 3 minutes. During any one session studying a single subject, a series of 2-5 runs was carried out consecutively according to time available, and/or to the likelihood of rejecting the first one or more runs on account of unsettled heart rate and ventilation. The scheme for a single session is shown in Fig. 5:11.

The subjects for these experiments were 5 healthy men (range 18 - 45; 18 - 23 except for subject M). Subject W was also in the first series. The total number of sessions was 14, and of runs, 38. The composition of these is shown opposite (Table 5:1).

EXTRACTION AND TREATMENT OF DATA

In general, most runs showed a clear increase in heart rate and usually also in ventilation, starting within a minute of the switch to 10% oxygen and usually levelling out before the end of the 3-4 minutes; reoxygenation caused the typical fall in ventilation within a few breaths, and usually a clear decrease in heart rate. An example of four serial sections of one run is shown overleaf (Fig. 5:12).

Fig 5:12



a,b,c,d: serial traces from same run. Top: CO_2 at the mouth; slight decrease during hypoxi and increase after first 2-3 breaths of oxygen(d). Middle : heart rate; increase during 2n min(b) and decrease after first 2 breaths of oxygen. Lower: $\dot{V}_I$(see text) and tidal volume; increase after 1 min and decrease at reoxygenation.

Records were examined and worked on in the following way:

A. Extent of changes in ventilation and heart rate

For 1) the final two minutes of air-breathing: reference period

2) the whole of the hypoxic period

3) the first minute of reoxygenation

the following variables were measured, and the values charted as shown in Table 4 overleaf:

1) For ventilation

a) Ventilation rate ($\dot{V}_I$) as defined (p.5/26) in arbitrary units, measured in mm deflection, for each breath.

b) Tidal volume (V_T) in those runs where this was recorded in addition to a). Also in arbitrary units, and for each breath.

c) End-tidal PCO_2 (P_ACO, torr) was derived for successive half-minutes of the periods concerned by taking mean plateau values drawn by eye over several breaths on the calibrated trace.

2) For heart rate

a) The lowest heart rate in a respiratory cycle, expiratory heart rate, (h'_E)

b) The highest heart rate in a respiratory cycle (h_I)

c) The amplitude of the respiratory oscillation in heart rate

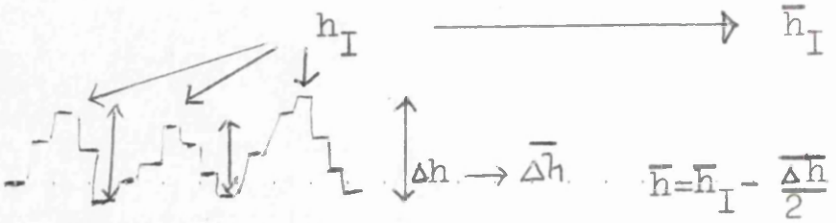
These three require further explanation:

Some authors have stated that respiratory heart rate rhythms cannot be defined at frequencies of breathing (f) higher than "very slow" (Davies & Neilson, 1967). In the present and other studies it was virtually always possible to find values in association with each breath for (i) the lowest heart rate during the preceding expiration and (ii) the highest during the inspiration, which were related to the breath in the timing appropriate to "inspiratory heart rate transients" as described by these authors and by Clynes (1960): that is, there was a rise in heart rate approximately 1 second after the start of an inspiration. It was hoped that by extracting this information, three purposes could be served; firstly to find out if h_I or h_E were an acceptable index of mean heart rate for these studies; secondly to see whether increasing heart rate could be attributed to either a greater inspiratory rise on the one hand, or a smaller expiratory fall on the other, and thus find evidence of predominantly either a cardio-acceleratory, or an anti-inhibitory mechanism; thirdly, to accumulate information for later analysis in a proposed study of sinus arrhythmia.

Table 5:2

Comparison of different indices of heart rate derived from traces

Three subjects, third occasions, three runs
(B, H, S, 3:1, 3:2, 3:3)



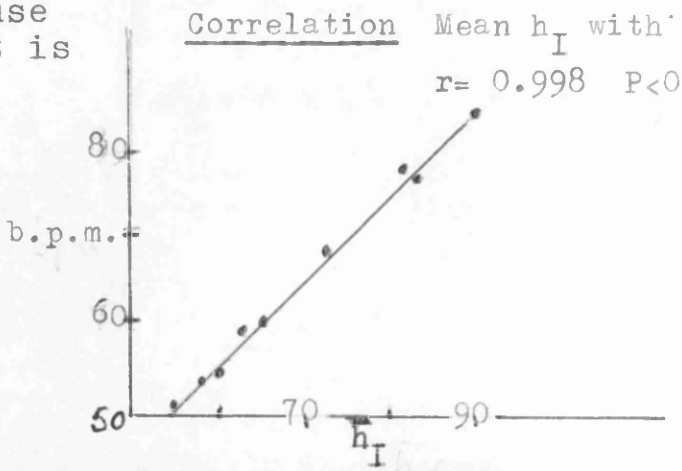
		Values for heart rate		
		$\bar{h}_I$	$\bar{h}$	Counting beats
B	Ref.	83	78	78
	H	90	86	85
	O	82	79	79
H	Ref.	55	52	51
	H	65	62	61
	O	57	55	54
S	Ref.	63	59	60
	H	73	68	69
	O	60	57	55

	Values for mean increase during hypoxia		
	$\bar{h}_I$	$\bar{h}$	Counting beats
B	7	8	7
H	10	10	10
S	10	9	9
Mean	9	9	8.6

	Values for mean decrease at reoxygenation		
B	-8	-7	-6
H	-8	-7	-7
S	-13	-9	-14
Mean	-9.6	-7.6	-9

The very slight over-estimation of increases and decreases by the use of h_I confirms that it is a suitable index for changes in heart rate

Correlation Mean h_I with beats/min:
r= 0.998 P<0.001



The first of these is of immediate importance, and will be expanded further: where a beat-by-beat record of heart rate is made, and shows fluctuations, it is necessary to decide on a method of measuring mean heart rate over a given period. Each pulse in that period can be counted, or a line drawn representing the mean of the oscillations; alternatively, if both h_I and h_E were to increase or decrease similarly, it would be possible to use one of them as an index of the change in mean heart rate. This would not be possible if the amplitude rather than the mean, were changing. The change in amplitude, h_I, h_E and beats/unit time were all measured in a few representative runs; it was possible to conclude that breath by breath h_I would give an appropriate index to mean heart rate changes (Table 5:2).

When values had been entered on a single-run data sheet (Table 5:3)

1) a graph was plotted (Fig. 5:13) from which timing information could be derived (period of steepest rise etc.).

2) the data were divided into the required periods, and the means, standard deviations and standard errors calculated for all variables for each period as follows:

i) Reference period

ii) Second and third minutes of hypoxia ($H_{2,3}$)

iii) Last minute of hypoxia (H_{Ω})

iv) First half-minute of oxygen

v) Second half-minute of oxygen

Instead of (iv) and (v) for later analyses it was realised that the first minute LESS the first ten seconds was more representative of the re-oxygenation changes, so a further period is defined

vi) Tenth to sixtieth sec of oxygen (O_{10+})

Statistical treatment for individual experiments

Student's t-test was applied to discover the significance of difference between means for these periods, for each run, for each subject individually:

Ref $\longrightarrow$ $H_{2,3}$:- an indication of the change due to hypoxia.

$H_{\Omega} \longrightarrow O_{10+}$:- an indication of the change due to reoxygenation.

These values, and significances, were recorded on a data sheet for the whole session of 2 or more runs (Table 6).

A full example of the treatment of data for a representative run and session follows in the next 4 pages (Table 5:3, Fig. 5:13, Table 5:4).

Table 5:3

Example of treatment of data - I

Data for subject H, session 2, run 3, as extracted from traces. (each value is for a single breath)

Air-breathing Ref

$\dot{V}_I$	h_E	h_I	Δh	
23	43	46	3	A
26	45	48	3	
25	45	47	2	
23	42	46	4	
25	44	47	3	
26	46	52	6	
26	48	48	0	
26	46	50	4	
27	49	51	2	
28	50	53	3	
27	48	52	4	
32	52	58	6	
28	52	53	1	
32	48	53	5	
39	50	56	6	
30	-	-	-	
30	52	57	5	

(Ref. n=17)

(Means etc. entered on session sheet) (overleaf - 5:14)

2nd and 3rd minutes of 10% O_2 i.e. starting at 1 min

31	44	47	3	H
29	45	47	2	
28	45	49	4	

Hypoxia - contd.

$\dot{V}_I$	h_E	h_I	Δh	
28	47	52	5	
27	47	48	1	
30	46	50	4	
28	48	50	2	
29	46	50	4	
28	47	52	5	
29	48	54	6	
28	50	54	4	
28	49	52	3	
30	49	56	7	
35	54	60	6	
36	54	56	2	
40	50	57	7	
35	54	62	8	
32	49	56	7	
30	50	57	7	2 min
28	54	54	0	
28	50	55	5	
30	52	59	7	
30	52	60	8	
36	51	61	10	
40	57	62	5	
39	57	66	9	
36	60	64	4	
33	52	57	5	
34	52	59	7	
29	55	60	5	

Hypoxia - contd.

$\dot{V}_I$	h_E	h_I	Δh	
27	52	57	5	
31	53	63	10	
34	61	64	3	
32	47	52	5	
31	48	52	4	3 min $H_{2,3}$ n=35
32	50	56	6	
30	57	62	5	
31	51	58	7	End H_{Ω} n=18

Reoxygenation

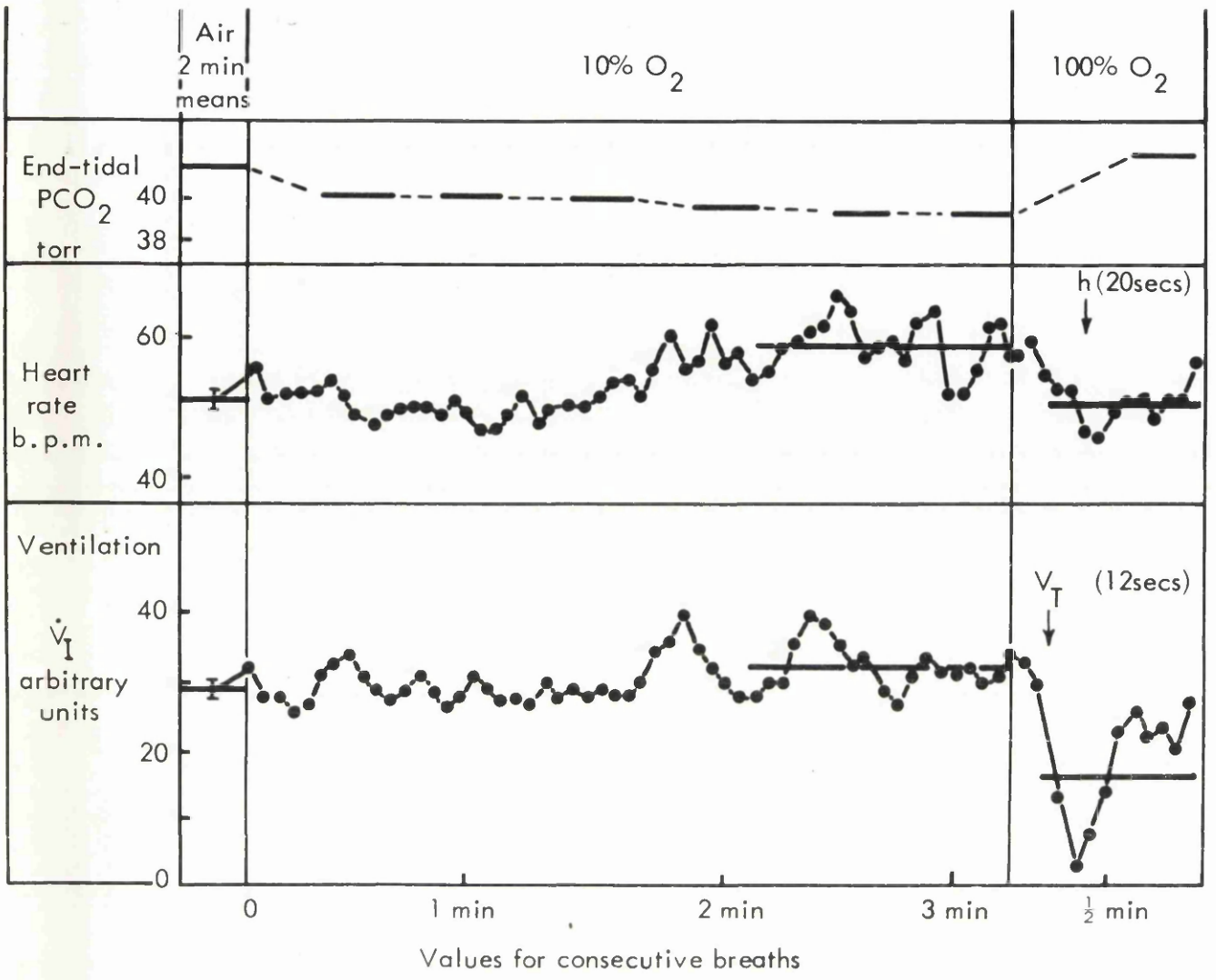
35	51	58	7	
34	58	60	2	
30	50	55	5	10"
17	51	53	2	
14	52	53	1	
3	46	47	1	
8	46	46	0	30"
14	46	50	4	
23	48	51	3	
26	47	51	4	
22	49	49	0	
24	48	52	4	
20	50	52	2	
27	53	57	4	1 min

1st 30" n = 7

2nd 30" n = 7

"O_{10}^{+}" n = 11

Example of treatment of data- II Fig 5:13 Subject H Run 2:3



Top: End-tidal PCO_2: each value a mean over 4-5 breaths

Middle and bottom: horizontal lines indicate mean values during

i) Reference air-breathing period of 2 min
ii) Last min breathing 10% oxygen
iii) 10th-60th sec after change to 100% oxygen

$V\downarrow, h\downarrow$: time of first clear fall in tidal volume and in heart rate indicated here, but noted from original trace.

Table 5:4

Example of treatment of data III

Subject H, Results for single session (2) - 3 runs

(Each figure is a mean of values for each breath in the period of study. Bracketed figure - S.E.M.)

	$\dot{V}_I$	h_E	h_I	Δh
		"R" Reference period		
2:1	33.56	50.94	55.39	4.44
	(0.77)	(0.52)	(0.66)	(0.63)
2:2	29.82	48.82	53.47	4.65
	(0.65)	(0.49)	(0.96)	(0.74)
2:3	28.82	47.50	51.06	3.56
	(0.95)	(0.81)	(0.97)	(0.45)
	"$H_{2,3}$" 2nd and 3rd mins of hypoxia			
	(Significant differences from Ref. period are underlined)			
2:1	34.63	57.14	64.74	7.57
	(0.63)	(0.61)	(0.51)	(0.3)
2:2	33.23	50.76	56.30	5.71
	(0.45)	(0.49)	(0.57)	(0.35)
2:3	30.64	50.63	55.83	5.06
	(0.98)	(0.65)	(0.88)	(0.41)
	"H_Ω" Last min of hypoxia			
2:1	33.22	57.33	64.56	7.17
	(1.03)	(0.85)	(0.71)	(0.37)
2:2	33.84	52.33	57.84	5.95
	(0.73)	(0.48)	(0.67)	(0.32)
2:3	32.39	53.28	59.28	6.11
	(0.84)	(0.95)	(0.97)	(0.49)

Table 5:4 contd.

	$\dot{V}_I$	h_E	h_I	Δh
	1st half-minute of 100% oxygen			
	(Significant differences from last minute hypoxia underlined)			
2:1	24.49	55.43	59.29	3.86
	(5.0)	(2.41)	(3.27)	(1.30)
2:2	22.90	52.67	56.33	3.67
	(5.87)	(3.04)	(3.25)	(1.99)
2:3	18.29	50.57	53.14	2.57
	(4.32)	(1.54)	(1.97)	(0.95)
	2nd half-minute of 100% oxygen			
2:1	19.10	51.50	56.00	4.50
	(3.92)	(1.98)	(1.75)	(2.29)
2:2	19.03	47.29	49.71	2.43
	(1.14)	(0.36	(0.57)	(0.61)
2:3	20.06	48.71	51.71	3.00
	(1.48)	(0.87)	(0.97)	(0.58)
	$\underline{O}_{10+}$ First minute of 100% O_2 excluding first 10 secs			
	(Significance of differences from last minute of hypoxia underlined)			
2:1	17.61		55.00	
	(2.88)		(1.28)	
2:2	16.44		49.70	
	(1.64)		(0.54)	
2:3	16.30		51.00	
	(2.03)		(0.91)	

--- $P < 0.05$

< 0.01

< 0.001

B. TIMING OF CHANGES IN VENTILATION AND HEART RATE

(1) During hypoxia

For each run, the values for heart rate and for ventilation were plotted for each breath (Fig.5:13). The resulting graphs were used to determine the period of steepest rise for each variable. A slope was drawn through this by hand, and the points at which it intersected the mean control level, and the mean level for the last minute of hypoxia (or the period during which a plateau level was maintained if shorter than the last minute), were taken as the beginning and end of the steepest response. These times were noted, and also in each case whether the start of ventilatory increase preceded or followed heart rate increase.

(The example in Fig.5:13 is one in which it was relatively easy to define the increase in heart rate, but not that in ventilation which in this instance was non-significant; note that there was nevertheless a marked reduction of $\dot{V}$ on re-oxygenation).

(2) At reoxygenation

All traces were worked over, adopting the following procedure:

i) Considering the heart rate alone, for each experiment, the re-oxygenation period was studied, and the first value found for either h_I or h_E which was lower than its range during the last minute of hypoxia. The delay was recorded between inspiration of oxygen and this first clear fall in heart rate

> A single value below the range of the hypoxic values, such as a bigger swing of heart rate in the course of one breath, would not necessarily indicate the beginning of the fall due to re-oxygenation. In practice, this "first fall" was always obviously the beginning of a continuing decrease in heart rate (e.g. Figs. 5:8, 12d).

ii) Tidal volume was next considered, and a similar procedure applied - the first distinctly smaller breath was found, and the time interval from the start of the first breath of oxygen to the start of this breath, was noted (e.g. Fig. 12d).

i) and ii) were carried out separately, without reference to each other.

Selection of results for analysis

The runs shown bracketed on Table 5:1 were not included in the initial analysis which comprised 30 experimental runs. For some results, 35 runs have been analysed, but criteria for inclusion in the main series were :

1) Complete air-hypoxia-oxygen data, with technically satisfactory continuous ventilation and heart rate records.
2) Absence of spurious changes in heart rate, e.g. due to interruptions at crucial moments, or changes clearly due to falling asleep.
3) Absence of any significant degree of "settling", that is a tendency for the baseline levels of heart rate and ventilation to fall between runs. (This latter proved not to matter - p. 5/55).
4) Assurance that hypoxia had in fact been induced. (Although P_AO_2 was not measured in every experiment, if the expected changes did not occur and a leak in the inspiratory supply was suspected, the end-tidal level was checked).

Table 5:5

Incidence of significant mean changes in last minute of hypoxia, relative to reference level

		$P < 0.001$	< 0.01	< 0.05	N.S.	Total
Number of instances in which significant increase occurred in	V	20	4	1	10	35
	h	31	3	-	1	35

Results

I. Individual experiments

(1) During hypoxia a significant increase in heart rate developed in 34 of 35 experiments. By comparison, the response of ventilation was less consistent; there was a significant increase in 25 of 35 (Table 5:5). Increases ranged from 2-17 b.p.m. for h, and from 0 to 40% Ref level for $\dot{V}_I$.

> Pattern of ventilatory change: a significant increase in $\dot{V}_I$, without one in V_T demonstrates a predominant increase in frequency of breathing - often stated to be the characteristic effect hypoxia. This in fact occurred in only 6 of 25 runs in which both parameters were recorded and the increase in V_I was significant.
>
> Pattern of heart rate change: there was an increase in the amplitude of the respiratory transient in only 6 of 30 runs, when the mean for reference period was compared with that for '$H_{2\cdot3}$'. That is, the pattern of heart rate oscillations remained unchanged whilst the mean level rose. (This implies that during the increase, there must have been either a greater inspiratory rise or a smaller expiratory fall, or both. Attempts to analyse this have been unrewarding).

The changes in end-tidal PCO_2 were of course dependent on the ventilatory change: the values were small, and did

Table 5:6

Control experiments with sham changes

Subject	Air	"Hypoxia" (H_{Ω})	"Oxygen" (O_{10-60})
	Ventilation V_I (arb. units)		
C	515	465	520
B	300	320	320
S	305	285	265
Means	373	357	361
	PCO_2 (to nearest 0.5 torr)		
C	39	39	38.5
B	39.5	40	39.5
S	40	39.5	39
Means	39.5	39.5	39.1
	Heart Rate		
C	61	61	62.5
B	84	82.5	80.5
S	63.5	65	60
Means	70	70	68

Each value represents mean from two runs

not exceed -4 torr in any single case: the calculated decrease in P_ACO_2 for a level of ventilation which is higher than baseline by 10% would be about 0.5% or 3-4 torr, so that on average, as well as in individual cases, the ventilation and CO_2 measurements were mutually confirmatory (Table 5:12, p.5/67).

End-tidal PO_2 measured on 12 occasions showed a mean of 44.7 torr, range 39.9 - 51.3. (The predicted P_AO_2 for 10% O_2 is 36-40 depending on R).

(2) At re-oxygenation

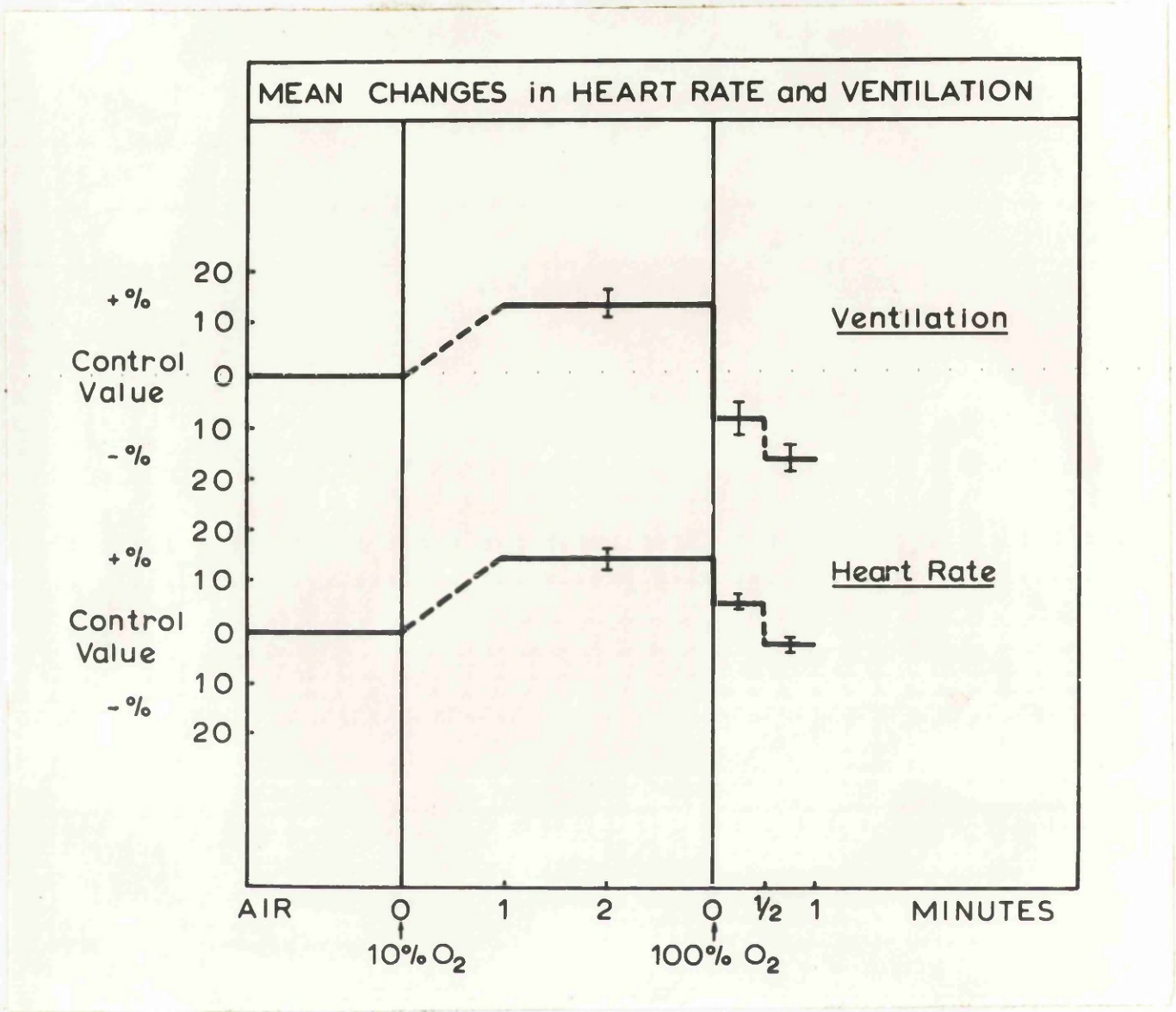
The change from the last min of hypoxia to the 10-60 second period after starting oxygen, showed significant decreases in heart rate in 26 of the 30 runs, and in ventilation in 27 of the 30. The extent of the change ranged from 0 to -16 beats per min and from -1 to -42% of the hypoxic level of ventilation ($\dot{V}_I$ decreased as low as -40% of control value).

The two changes sometimes appeared to be concurrent, but sometimes clearly not so, and the decrease in heart rate preceded that in ventilation as frequently as it followed it (Table 12b, p.5/71-72).

Control experiments

The mean values for heart rate and for ventilation during the standard measurement periods as for the air-hypoxia-oxygen experiments are shown in Table 5:6. There were no significant changes.

Fig 5:14



Mean values during i) second two minutes of 10% oxygen ($H_{2,3}$)
ii) first half-min after switch to 100% O_2
iii) second half-min after switch to 100% O_2
expressed as % ref (air-breathing) level.

Bars represent ± 1 S.E.

Ventilation is seen to decrease to well below control value, and heart rate almost exactly back to control value, on re-oxygenation.

II. Overall Treatment of Results A. Extent of Changes

(1) Comparisons of means

For the first main analysis of 30 runs, all were treated as equal whether from the same or different subject or session. Mean results were obtained and are shown graphically opposite (Fig.5:14). Tables of values are deferred to p.5/66 onwards, because the results will be shown compared with those of a further series still to be described.

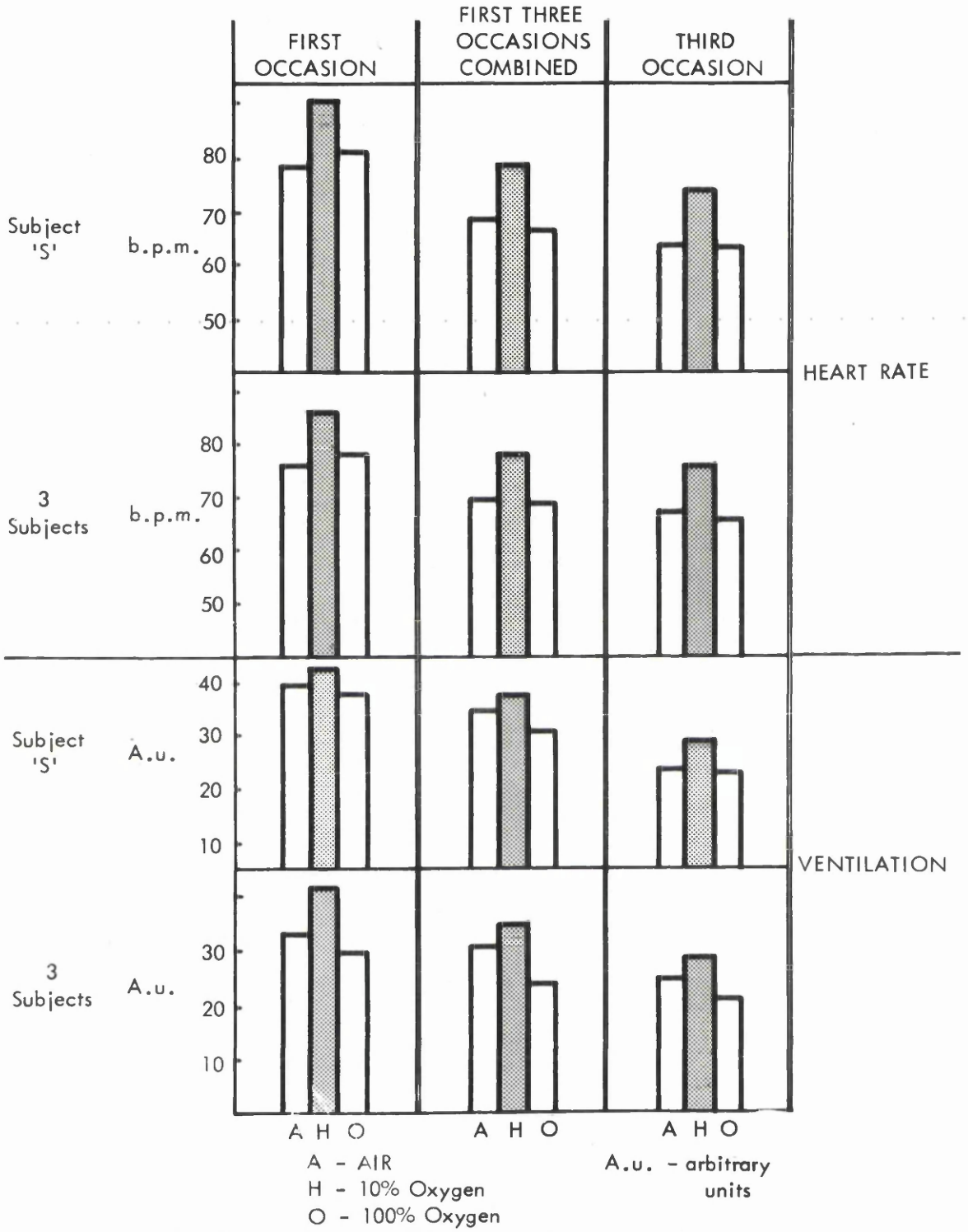
In summary, the mean increase in both heart rate and ventilation was by about 12% of the reference air-breathing level, during the 2nd-3rd minutes of hypoxia.

The decrease in ventilation on re-oxygenation was on average to 10 or 15% BELOW the reference level. The decrease in heart rate by contrast was to a level not significantly different from the reference level (Fig. 5:14) , and also compare ventilation with heart rate changes in Fig. 5:15 overleaf).

(2) Analyses of variance

In order to test more rigorously the significance of the results, and to define variations between different subjects, runs and occasions, analysis of variance was

Fig 5:15



Mean results from data used in analysis of variance; for significance of changes see text. This diagram shows that, although baseline levels become lower with training, the typical changes are seen even when the results for only one subject are considered, even when only one occasion is considered, and even when that is his first occasion.

carried out as detailed overleaf. The mean results included are shown opposite; three subjects were selected from the five because they were comparably studied on three occasions (c.f. Table 5;1, p.5/44). The conclusions from these analyses were as follows:

1. For heart rate

   In all analyses, whether for a single subject, or for three subjects combined, the effect of oxygen level on heart rate was highly significant, and independent of initial level, even though this varied greatly both within and between subjects. This was despite inclusion of first occasions, and first runs (Table 5;1).

2. For ventilation

   For three subjects combined, when the results obtained on their third occasions were considered, there was a significant effect of oxygen level on ventilation: when the first three occasions were combined, or first occasions only analysed, the variability in the effect on different subjects obscured any significant effect of oxygen level.

   Nevertheless, for single subjects, the effect

Table 5:7

Analysis of variance, three subjects

HEART RATE

(A) 1st three occasions

S.V.	D.F.	S.S.	M.S.	V.R.	P.
Subj.	2	7186	3593	39	<0.01
Occ.	2	1486	743	8	<0.05
O_2 level	2	912	456	33	<0.001
O_2X S	4	83	21	N.S.	
O_2 X Occ.	4	68	17	N.S.	
S X Occ.	4	365	91	6.5	<0.01
S X Occ. X O_2	8	52	6.5	N.S.	
Res-idual	27	388	14		
Total	53	10540			

(B) 3rd occasion only

S.V.	D.F.	S.S.	M.S.	V.R.	P.
Subj.	2	2909	1427	65	<0.00
O_2 level	2	306	153	7	<0.01
O_2 X S	4	18	4.5	N.S.	
Res-idual	9	256	28.4		
Total	17	3489			

VENTILATION

(C)

Subj.	2	367	184		
Occ.	2	836	418		
O_2 level	2	1047	524		
O_2X S	4	254	64	N.S.	
O_2 X Occ.	4	120	30		
S X Occ.	4	283	71		
S X Occ. X O_2	8	482	60	3.3	<0.01
Res-idual	27	470			
Total	53	3459			

(D)

Subj.	2	45	23	1.5	N.S
O_2 level	2	178	89	6	<0.02
O_2X S	4	37	9	N.S.	
Res-idual	9	159	18		
Total	17	419			

SV - Source of variation S.S. - Sums of squares V.R. - Variance ra
D.F. - Degrees of freedom M.S. Mean squares P - P value-significanc

of oxygen level was significant, and not variable between runs, or between occasions, even though the actual level of ventilation itself varied very considerably.

Details of analyses of variance

1. 3-factor analysis of variance. Three subjects, three occasions, three conditions of O_2 level (air, hypoxia, 100% oxygen. (Table 5:7). The first two runs for each of the occasions were used as replications. (First runs on any occasion, and first occasions, could be expected to be the least advantageous for demonstrating significant changes).

   Heart rate. The effect of oxygen level was highly significant ($p < 0.001$) - there was no significant interaction between subjects, occasions, oxygen level; heart rate varied significantly between subjects ($p < 0.01$) and between occasions ($p < 0.05$); subjects differed in the variability of their heart rate between occasions ($p < 0.01$) (Table 5:7A).

   (A 2-factor analysis for the three subjects, for their third occasions only, is included opposite (Table 5:7B) for comparison with the ventilation analysis, but was

necessary to establish significance; similar results were obtained using first occasions only).

Ventilation. The 3-factor analysis showed interaction between subjects, occasions and oxygen level (Table 5:7C). Separate 2-factor analyses for:

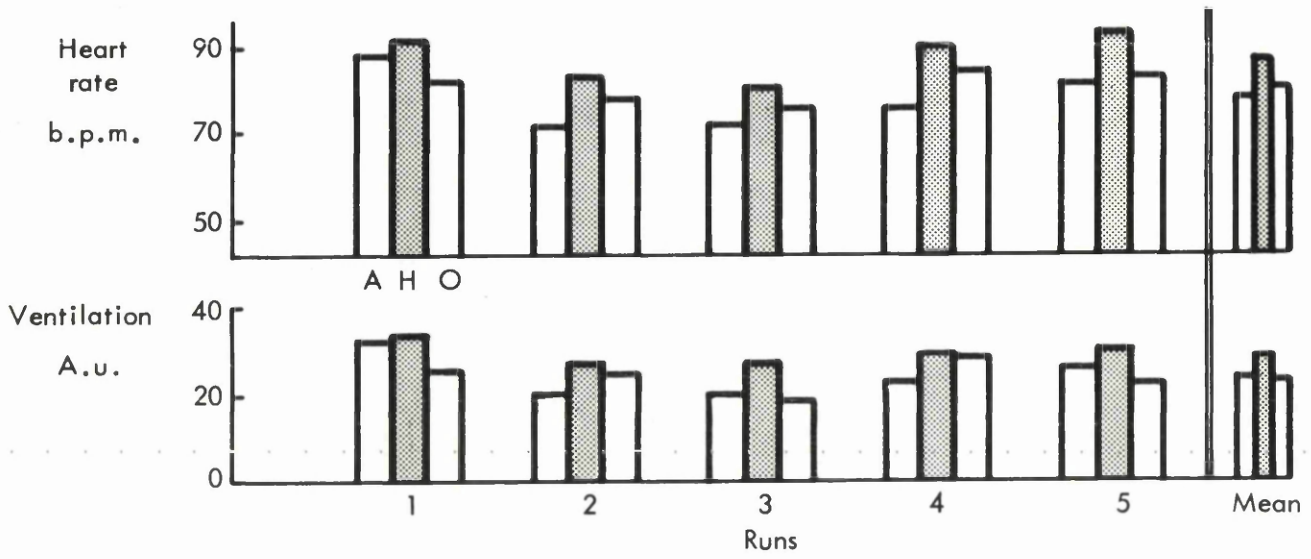
i) a single subject on three occasions showed a significant effect of oxygen level ($p < 0.001$) which did not significantly differ between occasions, although his ventilation did vary significantly ($p < 0.001$) showing a progressive decrease between successive occasions.

ii) all three subjects on their first occasion: the oxygen effect was not significant due to its variation between subjects (oxygen level: subject interaction $p < 0.05$).

iii) three subjects on their third occasion (Table 5:7D): the effect of oxygen level was significant ($p < 0.025$) and did not significantly vary between subjects.

A further analysis using three replications (1st 2nd and 3rd runs) for these same subject-occasions, improved the demonstrable significance of the oxygen effect to $p < 0.001$.

Fig 5:16



Results for one subject (S) on a single (3rd) occasion; 5 consecutive runs

Analysis of variance to demonstrate any variation due to the place of a run in the sequence of runs:

		Air → Hypoxia	Hypoxia → O_2
HEART RATE	Run effect (difference in h from one run to another)	$p<0.05$	$p<0.01$
	O_2-level effect (difference in h attributable to A → H , H → O)	$p<0.01$	($p<0.2$)
	Run x O_2 interaction (difference in effect of H or O on heart rate from one run to another)	NS	NS
VENTILATION	Run effect	$p<0.05$	($p<0.2$)
	O_2-level effect	$p<0.05$	$p<0.05$
	Run x O_2 interaction	NS	NS

It was confirmed that for heart rate the difference between air and 10% oxygen was significant, and the 100% oxygen values were similar to the air values; whereas for ventilation, the 100% oxygen values were lower than the air values.

2. 2-factor analysis of variance. One subject, 5 runs, two oxygen levels. Separate analysis for air→hypoxia, and for hypoxia → oxygen. The object of this analysis was to test for the significance of the effect of hypoxia, and of reoxygenation respectively, when a whole series of runs is included despite a high unsettled level in the first run.

   Heart rate: the effect of hypoxia in increasing heart rate was again shown to be significant ($p < 0.01$); that of reoxygenation in decreasing it on this occasion was not (p 0.05-0.2).

   Ventilation: the effect of hypoxia, and the effect of reoxygenation, were equally significant ($p < 0.05$).

Table 5:8

Correlations

	Between increases during hypoxia and decreases on re-oxygenation		Between changes in ventilation and changes in heart rate
For Ventilation	N.S.	Air ↓ Hypoxia	$r = 0.40$ $P < 0.05$
For Heart rate	$r = -0.72$ $P < 0.001$	Hypoxia ↓ Oxygen	N.S.

Correlations (Table 5:8)

It was of interest to know whether, during hypoxia, increase in ventilation was related to increase in heart rate, and whether at reoxygenation the decreases in either were related to their own increases during hypoxia.

The correlation coefficients were calculated between the appropriate sets of data and their significance found, with the following results:

1) Increase in heart rate was significantly correlated with increase in ventilation at the 5% level

2) Increase in heart rate during hypoxia was correlated at the 0.1% level with decrease in heart rate on reoxygenation (that is, heart rate returned to its original value)

3) Increase in ventilation during hypoxia was NOT correlated with decrease of ventilation on reoxygenation (that is, ventilation was depressed irrespective of the extent to which it had been stimulated)

This difference in behaviour of the two variables supported the concept of a different type of control mechanism.

B. Timing of Changes

(1) Changes during hypoxia

The mean time to the start of the steepest increase, and to the end of it, was found for ventilation

and for heart rate respectively. The means were compared and found not to differ significantly, but there was a tendency to an earlier and briefer rise in ventilation than in heart rate: ($\dot{V}$ 30-80 sec, h 50-120 sec).

Changes at reoxygenation

The intervals from the first inhalation of oxygen to the first clear fall in ventilation, and in heart rate respectively, were meaned and compared. There was no significant difference between them (Fig. 5:23 and Table 5:12, p<.71).

But these were mean results. The time intervals for ventilation and for heart rate respectively had been deliberately studied without reference to each other. When individual experiments were considered WITH reference to the relationship between the two responses it became clear that in some instances there was distinct dissociation, as already found in the subject K in the first series. These dissociations were to some extent evident in the treatment of data for individual experiments, in the mean values for ventilation and for heart rate respectively in the first, and second half-minutes of oxygen breathing respectively; thus in some instances, the

one had become significantly less than the hypoxic value in the first half minute, when the other had not. The information obtained in this way is set out in Table 5:12b, p.5/71.

Conclusions from this series

1) Heart rate increases significantly during three minutes of inspiring 10% oxygen, irrespective of subjects, and of any settling (between runs) or training (between occasions) effects.

2) Heart rate decreases within the first minute of reoxygenation to a level not significantly different from the initial air-breathing level.

3) Ventilation increases significantly during this same procedure, though there is more variation between subjects and between occasions for any one subject, than there is in the response of heart rate.

4) Ventilation is more constantly decreased at reoxygenation than it is increased during hypoxia, and this decrease is commonly to a level below the air-breathing level.

5) Ventilation and heart rate both decrease at reoxygenation within a timing similar to that found by other workers for ventilation, and attributed by them to a reflex involving peripheral arterial chemoreceptors.

6) The change in heart rate is not constantly concurrent with the change in ventilation, and precedes it as frequently as it follows it.

MAINTAINED VENTILATION EXPERIMENTS

Introduction

The next step appeared to be to find some method of separating the effect of reoxygenation on heart rate from its effect on ventilation. In man there are several possibilities for doing this:

1) To apply a similar routine during artifical ventilation

a) in anaesthetised patients.

This would be interesting as a comparison between awake and anaesthetised man, and between anaesthetised man and reported findings on anaesthetised animals. Ethical considerations make this type of experiment with 10% oxygen difficult.

b) in awake patients being ventilated artifically for reasons which do not imply abnormality of the lungs or of the control mechanism, such as respiratory muscle paralysis by motorneurone disease. Such an opportunity will be taken if it can be found.

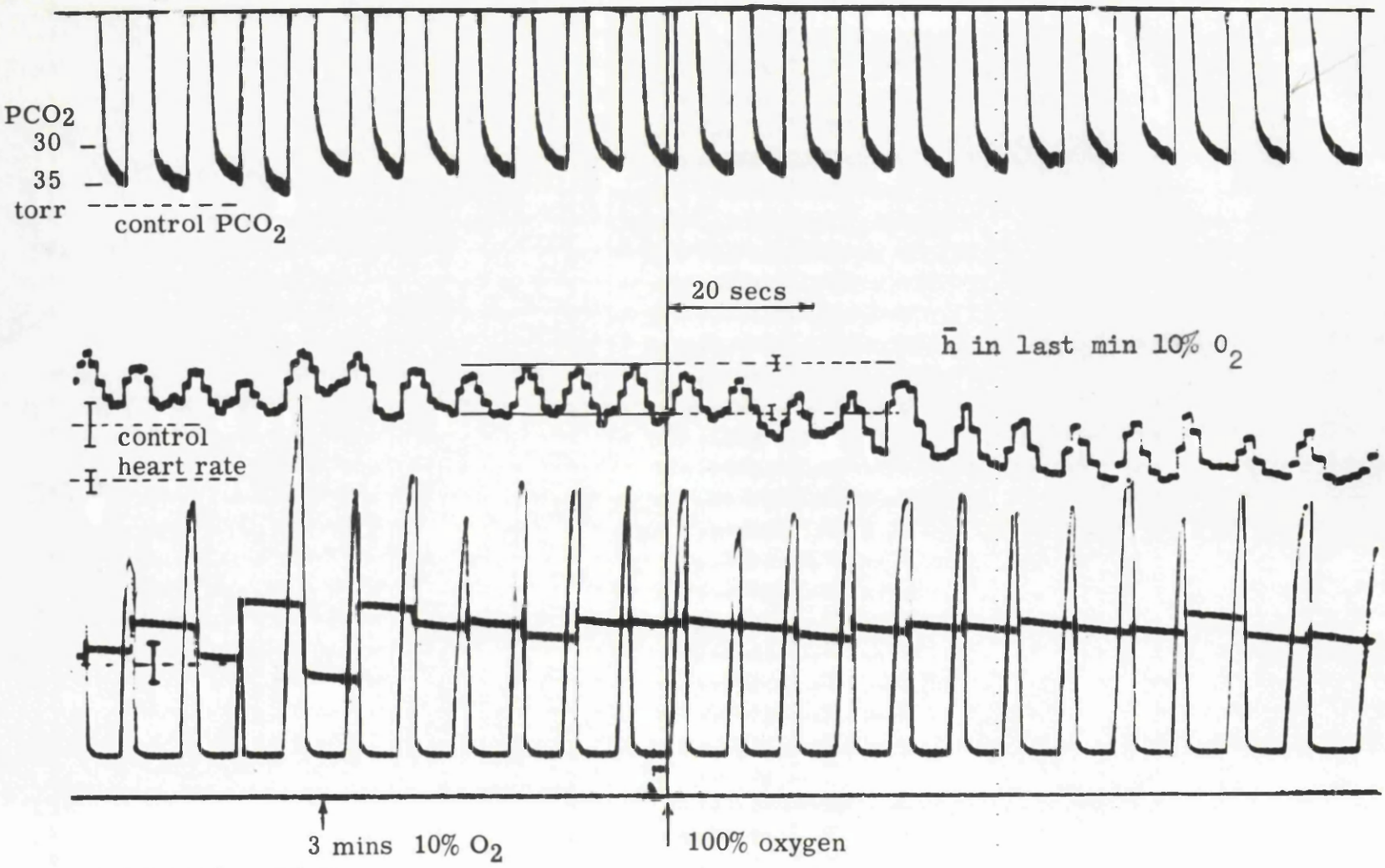
c) in curarised volunteers - not an acceptable procedure.

2) To apply a similar routine in awake normal subjects who could deliberately keep their ventilation steady during the changes in inspired gas. This was the method developed. It had the advantage that the ventilatory mechanics would be more normal than in artificial ventilation, but the possible disadvantage that the situation would be made abnormal, and perhaps the effect on heart rate invalidated, by the very fact of deliberate effort.

Method

Three of the subjects of the previous experiments were instructed in a method devised for maintaining an existing pattern of breathing. Pneumotachography was employed as described, and the pattern was displayed on a C.R.O. from the tidal volume output. The time base was adjusted according to the subject's respiratory frequency, so that each successive breath started at the same point on the screen. This was done with the subject NOT watching the screen; the procedure was straightforward only if the breathing pattern was regular. When a regular pattern had been established, the subject was asked to watch the screen and to maintain the same pattern.

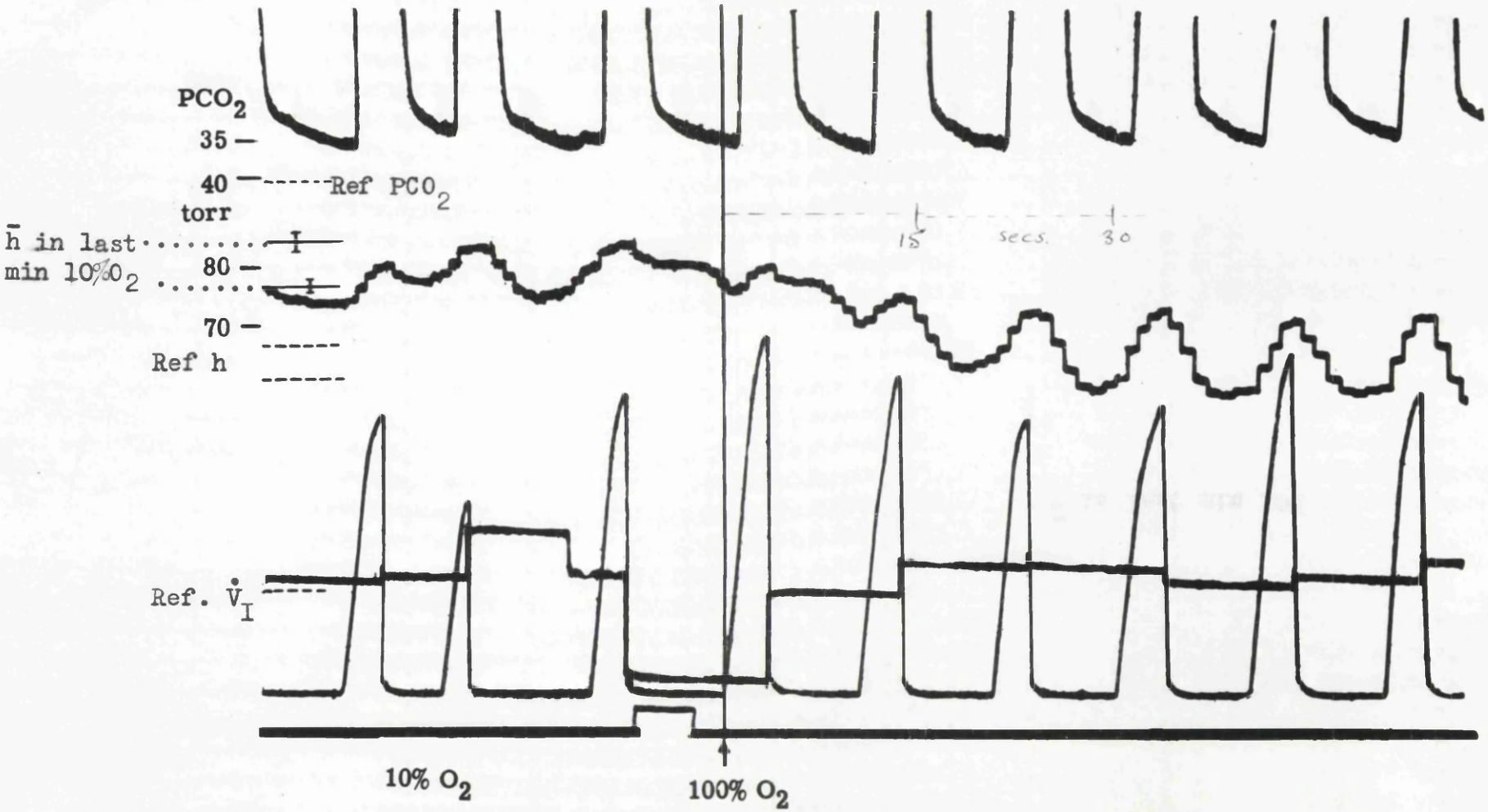
Fig. 5:17



Example of maintained ventilation over the period of the change from 10% to 100% oxygen. The subject hyperventilated, but a clear decrease in heart rate is shown.

Fig. 5:17

Fig. 5:18



A further example of maintained breathing pattern, showing an abrupt decrease in heart rate following re-oxygenation.

The initial tendency was to overventilate at the command to watch, and thereafter to maintain a fairly steady, but excessive, level (Fig. 5:17) but the subjects improved with practice. The method was further improved by using a storage oscilloscope - with this the pattern of the breaths in the required period could be stored so that there was no image memory required for the subject to start his breath at the correct point and achieve the correct tidal volume.

When the subjects were trained, the hypoxia-reoxygenation routine was carried out as before. The pattern was "stored" towards the end of the third minute of breathing 10% oxygen; by the time the adjustments were made, the command to watch and start "maintained ventilation" was usually at about 3½-4 min. After a few breaths, either (1) the change to 100% oxygen was made OR (2) a sham change was made to a similar hypoxic mixture, followed after half a minute by a change to 100% oxygen.

Results

A total of 16 such experiments was carried out, with more or less success. Some runs were excluded on the grounds of (a) failure to keep a steady maintained level over the crucial period, (b) excessive over-ventilation

Fig. 5:19

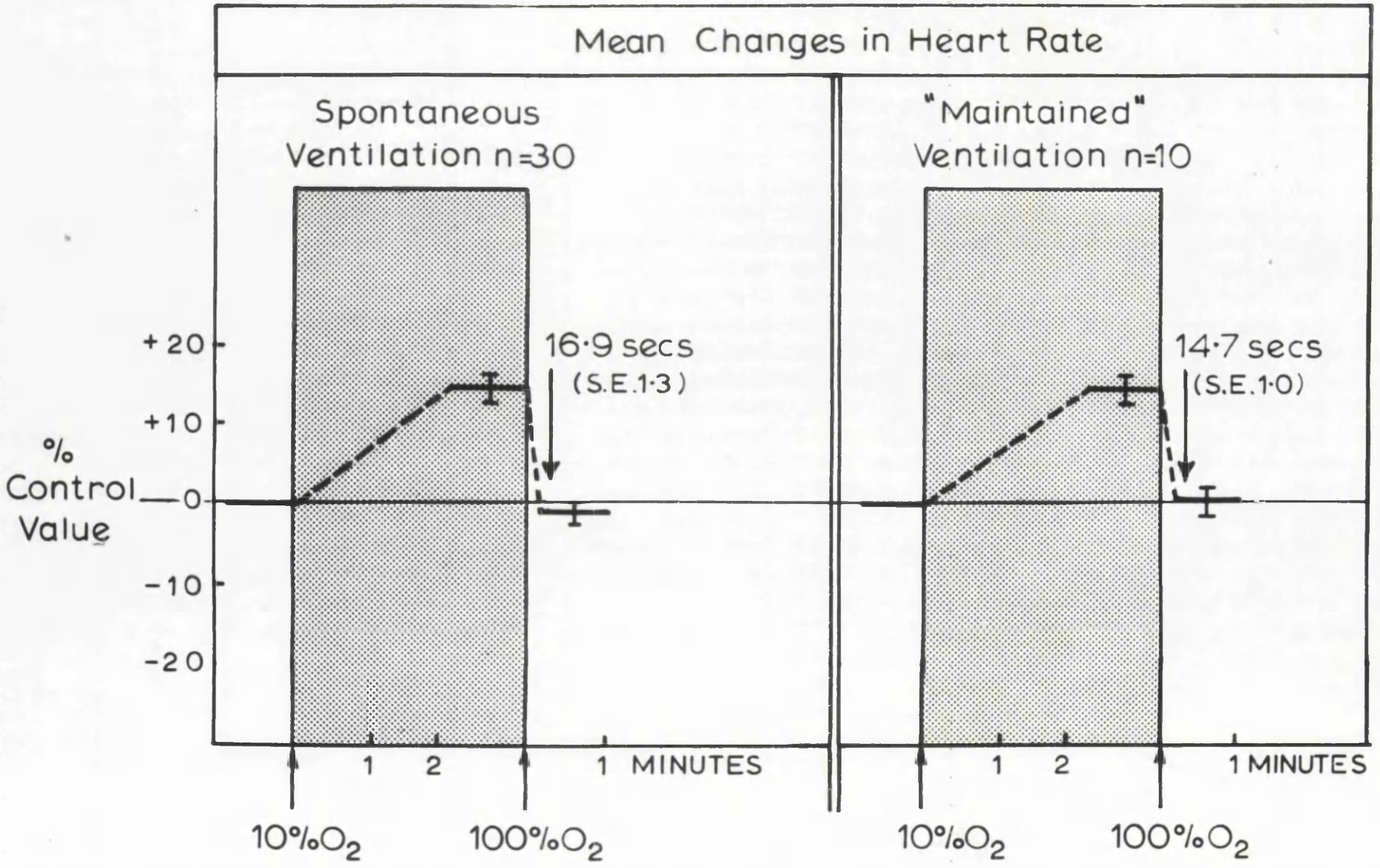


Fig. 5:19

Values for % change in heart rate are shown for the last minute of hypoxia, and the 10th-60thsec after the switch to 100% oxygen. Bars represent ± i S.E.M. change. Arrows indicate delay time to first clear fall (see Fig. 5:23). Heart rate changes are shown to be almost identical whether or not the breathing pattern is maintained.

during the maintained period (although not comparable with the "spontaneous" experiments these nevertheless gave results of interest in that the breathing was not allowed to decrease at reoxygenation) (Fig. 5:17), (c) spurious changes in heart rate during the watching or sham change. This left 10 runs for comparison with the "spontaneous" series for the timing data, and 8 for comparable experiments for the extent of the increase and decrease in heart rate and ventilation. Traces were examined and data extracted and treated in a similar way to that described for the "spontaneous" series.

Examples of traces from such experiments are shown in Figs. 5:17, 18, 21.

Comparison with spontaneous (second) series

Mean results, treating all runs as units irrespective of subject and occasion, were used in the main comparison with the "spontaneous" results, and these are shown in Tables 5:9, 10 overleaf and Figs. 5:19, 22 for extent of the changes, and Tables 5:12a,b, Fig. 23 for the timing of the changes, p.70-71.

In summary, the increases in heart rate and ventilation were not significantly different in the two series: the decrease in heart rate on reoxygenation was

Table 5:9

Changes in ventilation and heart rate during hypoxia and re-oxygenation

		Mean values in different periods expressed as % reference (air-breathing) values			
		Spontaneous breathing n = 30		Maintained breathing n = 8	
	Ref. value	100	S.E.	100	S.E.
Heart rate	10% O_2 2nd/3rd min	112.1	± 1.45	110.8	± 2.5
	10% O_2 last min	114.6	± 1.4	115.5	± 4.3
	100% O_2 first ½min	106.5	± 1.5	111.8	± 3.6
	100% O_2 10-60 sec	98.0	± 1.2 ← N.S. →	101.4	± 3.9
Ventilation	Ref. value	100		100	
	10% O_2 2-3 min	113.3	± 3.1	116.2	± 4.7
	10% O_2 last min	113.3	± 2.7	117.1	± 4.3
	100% O_2 first ½min	91.4	± 3.3	105.1	± 5.1
	100% O_2 10-60 sec	83.8	± 5.0 ← P<0.05 →	105.8	± 5.2
	% fall: last min 10% O_2 ↓ first ½min 100% O_2	-19.1	← P<0.05 →	-7.1	

Table 5:10

Changes in end-tidal CO_2 concentration during hypoxia

	Spontaneous			Maintained	
Change in % CO_2 at:	1 min	2 min	3 min	at 3 min	in last min
Mean	-0.16	-0.29	-0.37	-0.3	-0.4
range	0 to -0.3	0 to -0.4	0 to -0.5	0 to -0.5	-0.1 to -1.0
	(i.e. changes of the order of 2-3 torr)				

Table 5:11

Spontaneous v. Maintained breathing

Data used, and results of analysis of variance, for heart rate of one subject (S), comparing the effect of spontaneous and maintained ventilation on the response to hypoxia and reoxygenation.

(4 runs of each type used as replications)

	Air	Hypoxia	Oxygen
Spontaneous (S.V.)	(1) 56) (2) 65) 66 (3) 71) (4) 72)	67) 82) 78.5 88) 77)	50) 64) 60 71) 54)
Maintained (M.V.)	(1) 70) (2) 68) 64.5 (3) 58) (4) 62)	83) 71) 75 76) 71)	70) 62) 62.5 62) 56)

Each value is itself a mean over periods Ref, H_{Ω} and O_{10+} as defined (p. 5/48)

Source of variation	Degrees of freedom	Sums of squares	Mean square	Variance ratio	Significance (p)
$\underline{O_2}$ level	2	1068	534	28	< 0.05
SV/MV	1	3	3	<1	NS
Vent.XO_2	2	38	19	3	0.10 - 0.05
Residual	18	95	5.2		
Totals	23	2067			

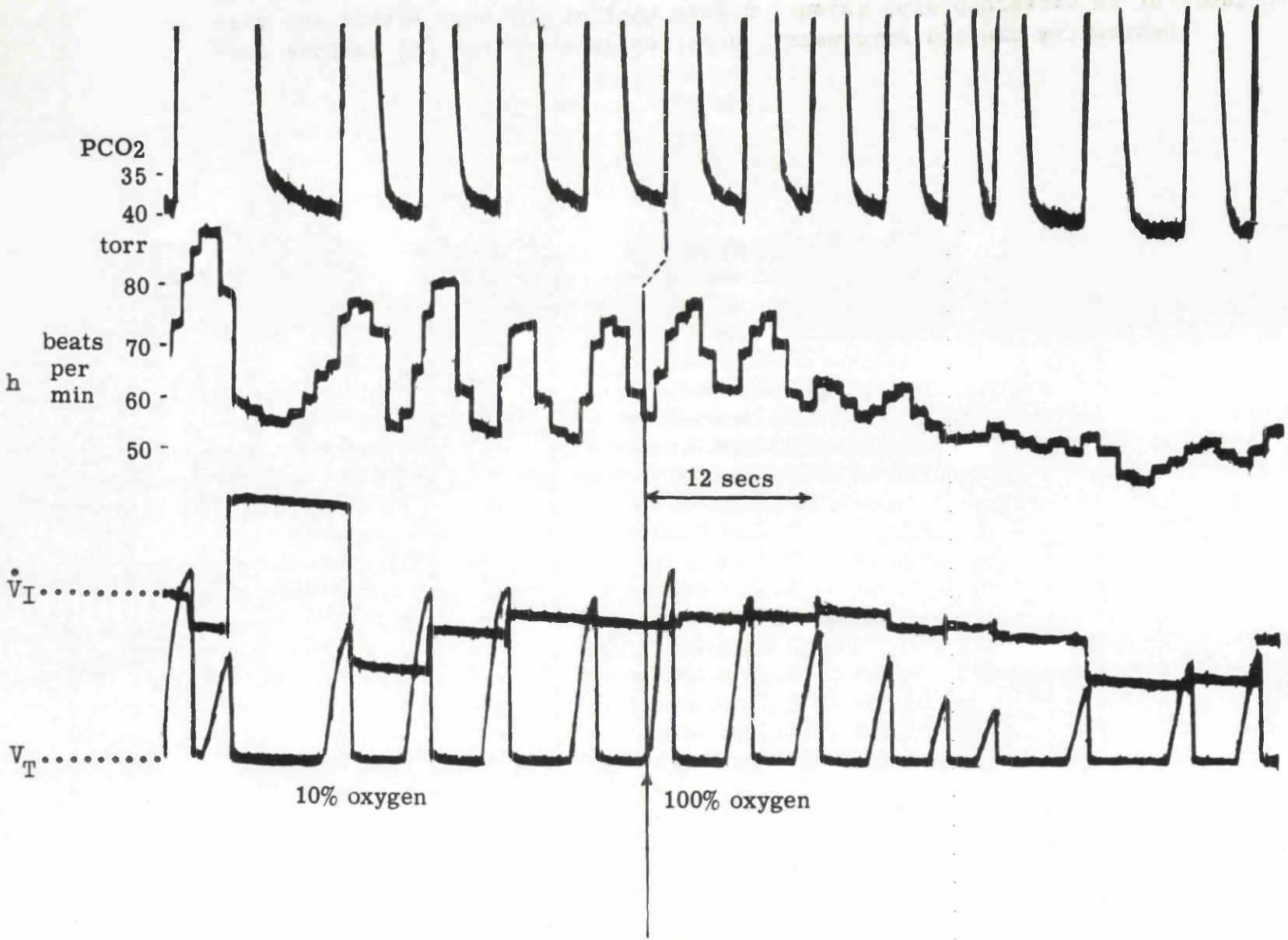
similar both in magnitude and in timing whether the breathing was maintained or not.

A more rigorous test of the comparison between the two ventilatory conditions was afforded by a two-factor analysis of variance on the results of a single subject (S) for four 'spontaneous' and four 'maintained' runs. (An example of one of each type on this same subject is shown in Figs. 5:20, 21). There were differences in the effect of oxygen level on heart rate with the different ventilatory conditions, but nevertheless there was a significant ($p < 0.05$) effect./ Table 5:11 When the change from hypoxia to oxygen only was analysed, the difference between the hypoxic effect in two conditions was not significant, and the effect of oxygen level highly so ($p < 0.001$) (Table 5:12).

Discussion

There were two possible sources of error which require comment:

1) The viscosity change between 10 and 100% oxygen was not allowed for in the system for maintaining ventilation: in all calculations of the tidal volume and ventilation rate during 100% oxygen breathing, a correction factor of 0.9 was normally



Subject S : The change from 10% to 100% oxygen, showing concurrent decrease in heart rate and in ventilation.

Fig. 5:20

Fig 5:21

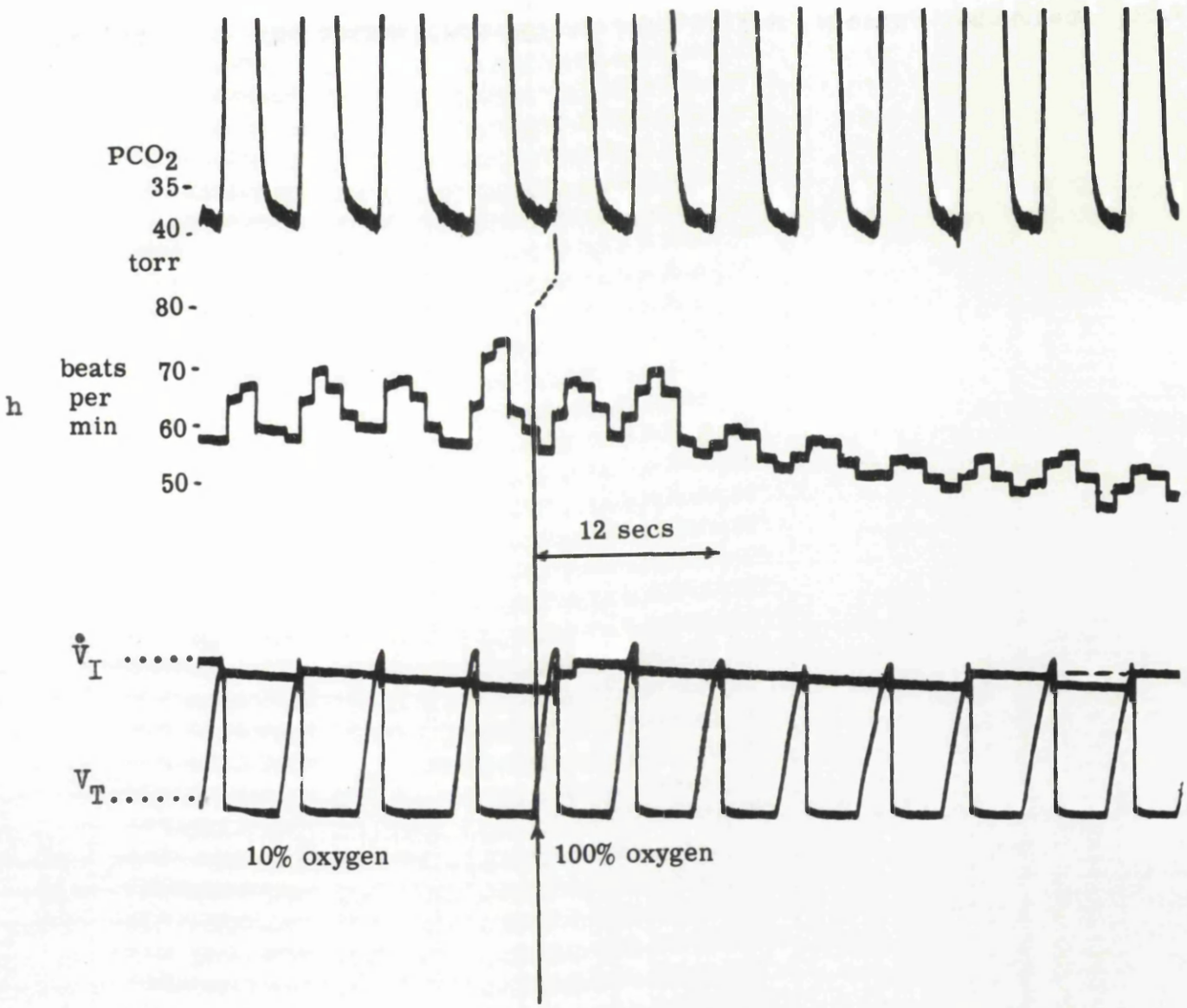
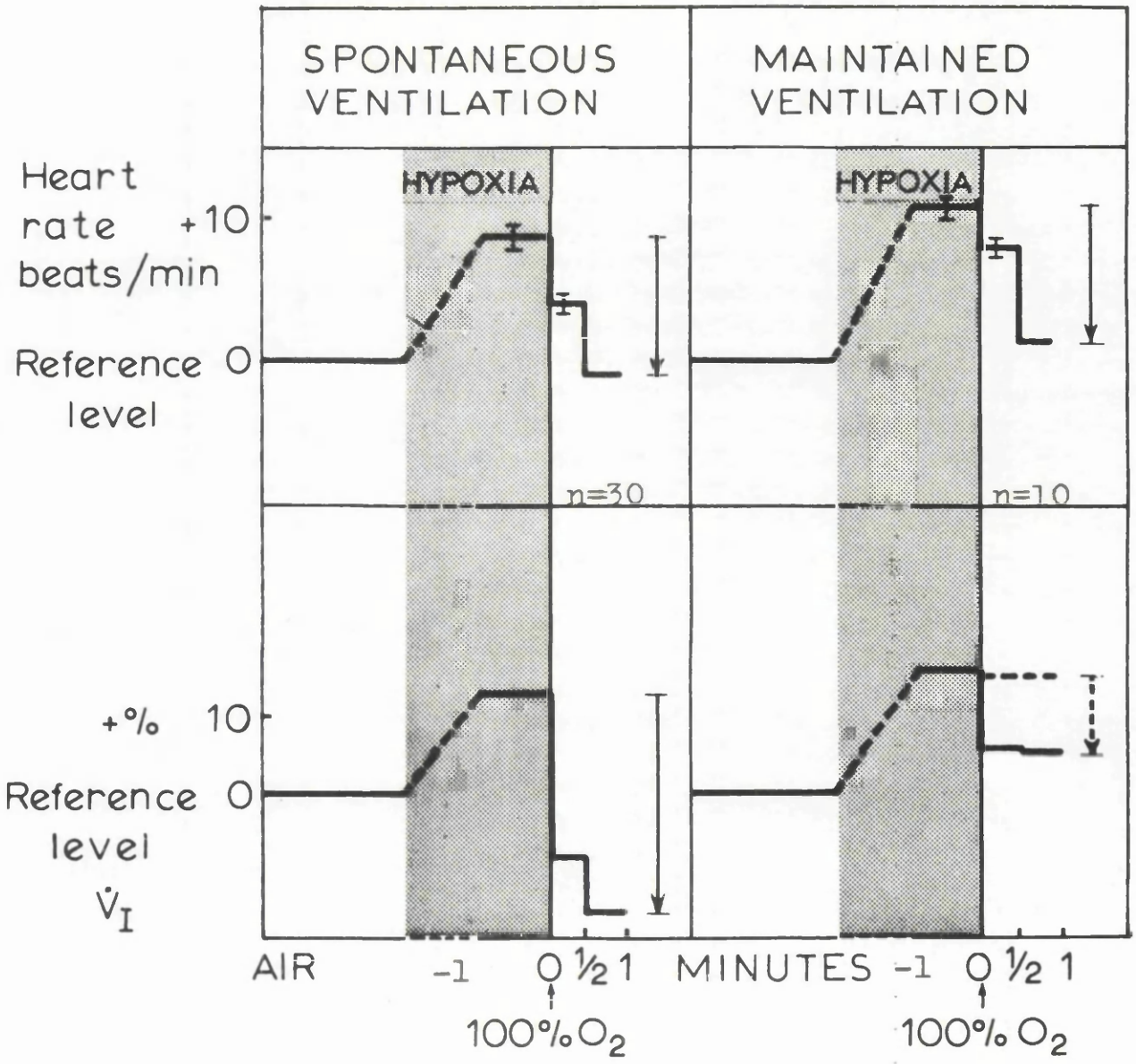


Fig. 5:21

Same subject (S) and occasion as above : breathing pattern maintained over the change from 10% to 100% oxygen ; heart rate decreases as in spontaneous breathing.

Fig 5:22



Changes at reoxygenation : comparison between spontaneaous breathing, and deliberate maintenance of pattern during the change from 10% to 100% oxygen.
Lower right: broken line shows apparently maintained V level; solid line, after correction for viscosity (see text)

applied to allow for the increase in viscosity (and therefore increase in apparent flow, and overestimation of tidal volume (see p.2/3 Fig. 2:2) During maintained ventilation no allowance was made for this: the subject maintained the same tidal volume as read by the pneumotachograph, and was therefore unknowingly aiming to breathe 10% LESS at the change to oxygen, when he kept an apparently steady level. The corrected change in ventilation is shown by solid line in Fig. 5:22 (opposite) and the apparent continuation as a broken line. This detracts from the value of the method, and, in retrospect, a compensation for this should have been built in to the system. However: (1) the calculated decrease in ventilation when apparently maintained, remained significantly less than the decrease during spontaneous breathing, whilst the decrease in heart rate was similar, and (2) the viscosity change was a step change at the first breath of oxygen: if ventilation were perfectly (apparently) maintained there would therefore be a 10% reduction in the first breath of oxygen. The decrease in heart rate was at 14-15 seconds so could not be attributed to this.

(2) Using the "h_I" as the index of heart rate: this has already been discussed in relation to the main series (p. 5/17.). Its use may overestimate the decrease in heart rate on reoxygenation, if there is abrupt reduction in respiratory oscillation. In the 'spontaneous' series this was shown to make no significant error in the results. If ventilation is maintained, and if this leads to a continuation of a similar oscillation of heart rate at reoxygenation, then, the measured decrease in heart rate would reflect the mean more closely than in the 'spontaneous' condition. On average, if the real mean reductions were the same in the two series, spontaneous and maintained, the tendency would be to produce results showing a falsely greater fall in spontaneous than in maintained ventilation. Since the point at issue is to demonstrate whether heart rate falls as much during maintained as during spontaneous breathing, this tendency would work against a significant result. The two ventilatory conditions have been shown to be accompanied by statistically similar falls in heart rate. The fact that h_I was used makes this result all the more significant.

Further details of the results for the 'spontaneous' and the 'maintained' series are given in the succeeding pages.

Table 5:12 a

Time course of changes in ventilation and heart rate

1. During development of hypoxia (35 experiments)

Period of steepest increase
(secs from first inspiration)

	beginning of increase	end of increase	
$\dot{V}$	31.0 ± 5.8	81.5 ± 15.1	n = 30
	N.S. ↕	N.S. ↕	
h	48.7 ± 8.5	119.6 ± 20.8	n = 35

Incidence of precedence

	No. of instances
$\dot{V}$ rose before h	19
h rose before $\dot{V}$	6
simultaneous rise	5
no rise in $\dot{V}$	5
	35

Table 5:12 b

2. At re-oxygenation

	Spontaneous breathing	Maintained breathing
	n = 30	n = 10
	Mean delay time before reduction (secs)	
V_T	16.4 ± 1.6 (S.E.M.)	-
h	16.9 ± 1.29 ← N.S. →	14.7 ± 1.04
	Incidence of precedence	
$V_{\downarrow}$ before $h_{\downarrow}$	13	-
$h_{\downarrow}$ before $V_{\downarrow}$	14	
simultaneous	1	
No clear fall in V	2	
	Pattern of change in heart rate	
h_I reduced before h_E	18	5
h_E reduced before h_I	12	5

Conclusions

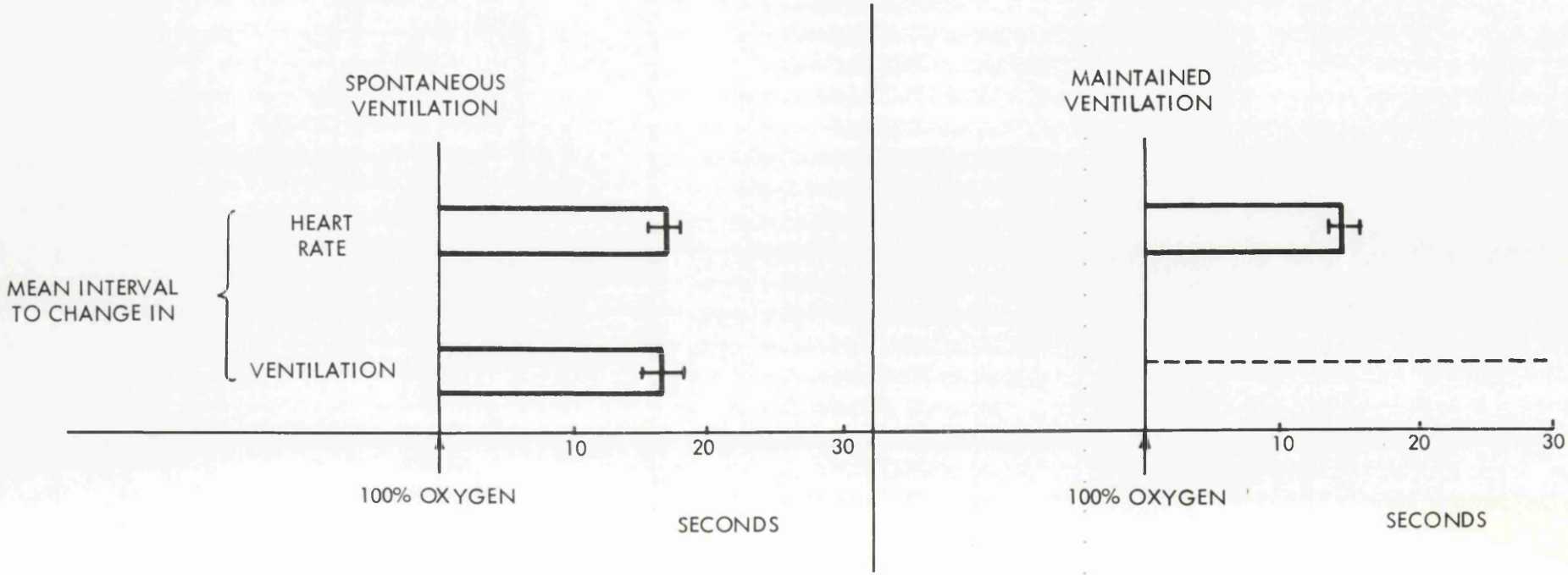
The exclusion of ventilatory change over the period of the change in heart rate makes it possible to conclude that:

(1) the tachycardia of hypoxia is reversed at reoxygenation independently of the reduction in ventilatory volume.

(2) this reversal takes place after a time delay which is at least as short as that for depression of ventilation.

Fig. 5.23

Re-oxygenation after 3-4 minutes hypoxia : delay time to the decrease in heart rate and in ventilation.



In spontaneous breathing, heart rate and ventilation decrease after a similar delay.

(Bars represent ± 1 S.E.M.)

When the breathing pattern is deliberately maintained, there is a decrease in heart rate after a similar delay to that in spontaneous breathing.

DISCUSSION OF THE SIGNIFICANCE OF THE TIMING OF RESPONSES AT REOXYGENATION(Fig. 5:23)

The delay time in all three series of experiments of 14-16 seconds from the beginning of the first breath of oxygen to the first clear reduction in ventilation, seems on the face of it a relatively long estimate of lung to chemoreceptor circulation time, plus reflex response time. It is, however, consistent with the findings of Dejours et al already referred to, and for which they presented good evidence, that this pattern of response was indeed attributable to chemoreceptor activity. Their timing was 10-13 secs measured from the beginning of the first breath of oxygen. The slightly longer time in the present series may be attributable to (1) slightly greater inspiratory instrumental dead space and/or (2) slightly different criteria for assessment of the first decrease. Certainly the pattern of response in the present series was essentially the same as that attributed to arterial chemoreceptors, and the repeatability of the timing assessed by the present method confirms the likelihood of this mechanism.

The time is taken up by lung ⟶ chemoreceptor circulation time, and the time necessary for oxygen to reach

alveoli such as to raise the mean alveolar tension above stimulus level.

In separate studies with CO_2 (see p. 3/10-11) it could be shown that with an average tidal volume the new gas at a step change would reach the mouth after about $\frac{1}{4}$ of the tidal volume had passed through: the dead space from the new gas front to the mouth was 100ml; it could be expected that of the "first breath of oxygen" reaching the alveoli (say 350ml total, a 500ml breath minus 150ml anatomical dead space), the first 150ml would be dead space gas (say 7% oxygen), the next 100ml 10% oxygen from instrumental dead space, and the final 100ml, 100% oxygen. If the FRC* were 2.5 litres, this represents an alveolar mixture at the end of the first breath of

$$\left.\begin{array}{rr} 0.15 + 2.5\ \text{L} & 7\% \\ 0.1\ \text{L} & 10\% \\ 0.1\ \text{L} & 100\% \end{array}\right\} \text{giving } 0.4\ \text{L } O_2 \text{ in } 3\ \text{L} = \text{about } 13\% \text{ oxygen, less some for transfer}$$

i.e. the alveolar PO_2 would still be below the normal air-breathing level. At the second breath a similar calculation gives a value of just under 17%, or 120 torr - sufficient to inhibit chemoreceptor activity. The time at which arterial blood would start to leave the

* Functional residual capacity

lungs hyperoxygenated would therefore be not sooner than the end of the "second breath of oxygen". At a 'typical' respiratory frequency of 12 per min with 500ml V_T this would make the apparent 15 sec delay time represent an alveolar-gas-change ⟶ ventilatory response time of not more than 8 sec. Given that the heart rate response occurred also after a delay time of 8 or less seconds, how could this throw evidence on the possible sites of action ?p.

1) Carotid chemoreceptors, by definition, when comparing ventilatory response, are a possible site.
2) Any site NEARER the lungs than this is possible, viz.
    a) coronary distribution - SA node
    b) aortic bodies
    c) pulmonary circulation - relaxation of hypoxic vasoconstriction might occur in similar timing.
3) Any site further away from the lungs becomes progressively less likely, but must be considered:
    a) medulla; it would seem likely that there would be only a marginal time difference between reoxygenation in carotid bodies, and in the medulla. However, if the timing of an

effect of CO_2 on the medullary chemoreceptors is any guide to the timing of the effect of oxygen on the cardio-acceleratory centre (and it may not be) there is evidence of a difference in timing (Lambertsen, Gelfand & Kemp, 1965). If oxygen is acting here on heart rate, then the other effects of sympathetic stimulation should be reversed in similar timing: in man this would be difficult to demonstrate since there is no marked vasoconstriction and rise in blood pressure, in this degree of hypoxia. (In the dog, this would mean one would expect to find a fall of blood pressure at the same time as the fall in heart rate. Neil (1956) found that the B.P. change occurred sooner).

b) peripheral haemodynamic effect. Conceivably a rise of blood pressure due to a reversal of local hypoxic vasodilatation in skeletal muscle, might lead to bradycardia on reoxygenation. This seems likely to take longer. Muscle blood flow studies are planned to investigate the timing of this.* It is also hoped that it will be possible to record blood pressure intrarterially in human subjects during

* Black & Roddie (1958) show a graph which suggests a greater delay in forearm blood flow reduction than in heart rate reduction, at the end of a period of hypoxia.

similar hypoxia-reoxygenation, to elucidate the order of events.

Further possibilities for investigation of this problem

It seemed that this was as far as the problem could be followed with the available subjects and equipment.

Table 5:13

Details of spinal patients studied

No.	Age	Level of lesion	Time since injury	Resting heart rate	
				Sitting	Lying
1	24	C7 complete	8m	60	60
2	19	C6 "	9m	80	71
3	36	C6 "	16yr	66	75
4	24	C7 "	$2\frac{1}{2}$m	123	78
5	20	complete D2 incomplete C4	4m	75	51
6	46	C7 complete	7m	75	73
7	40	C6 "	13yr	69	81
8	60	C7 "	8m	97	70

STUDY OF THE EFFECT OF HYPOXIA AND REOXYGENATION ON HEART RATE AND VENTILATION IN SUBJECTS WITH CERVICAL SPINAL TRANSECTION

Introduction

In studies of the causation of tachycardia in hypoxia the weight of evidence from animal experiments is in favour of a central action and a sympathetic efferent pathway; the only human evidence - that of the effect of beta-blocking drugs, is equivocal (p.5/12).

In patients with cervical spinal transection, there would be no possibility of centrally acting stimuli affecting the heart rate via the sympathetic (unless the spinal cord rather than the medulla were the reflex centre or site of direct action). Any effect mediated by the vagus, would, of course be intact.

It seemed of interest to carry out exactly similar experiments in such subjects, and this was arranged at the National Spinal Injuries Centre, Stoke Mandeville.

Subjects and methods

8 male patients were studied. All were known to have had signs and symptoms consistent with complete

PLAN OF AN EXPERIMENT

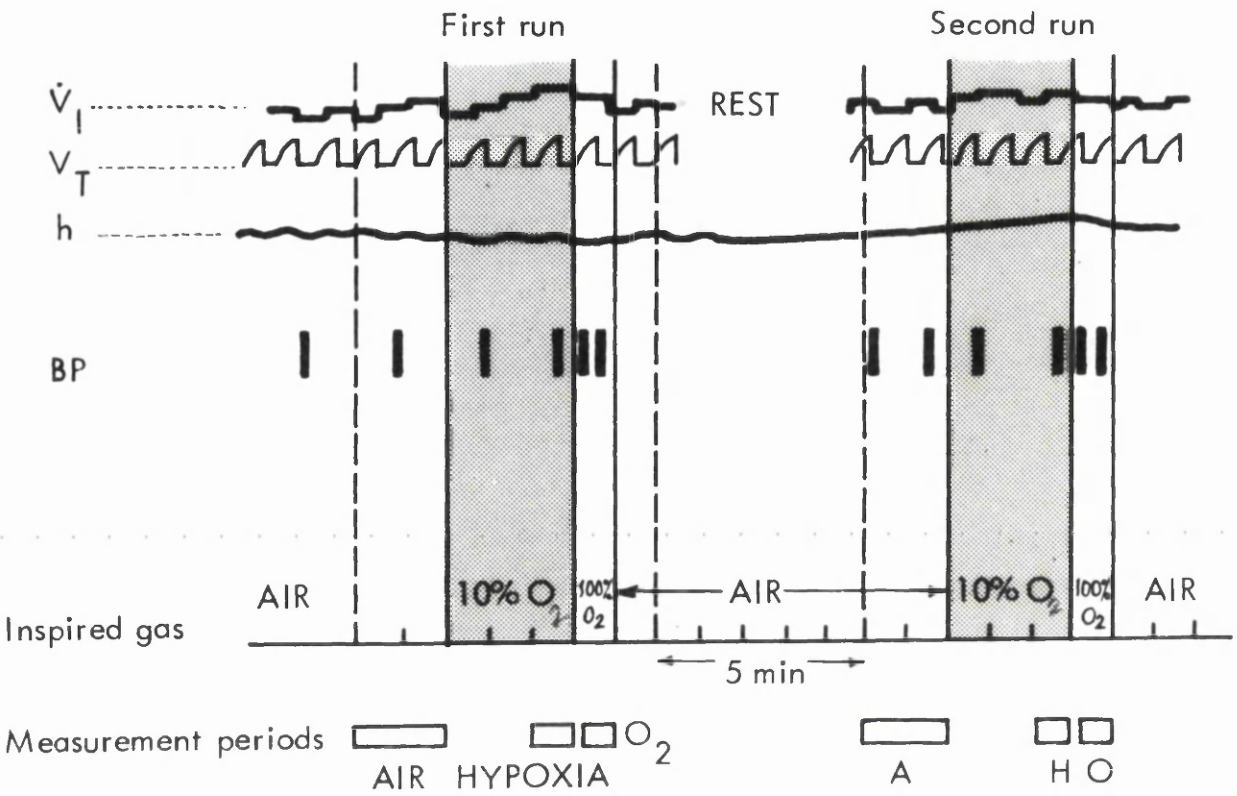


Fig 5:24

Spinal series

spinal transection above D2 for periods ranging from 2½ months to 16 years. Their age range was 19-60. All were in a good state of general health. Details are given in Table 5:13

Each subject was studied on two separate occasions within one week: once sitting in his wheel chair, and once lying supine in bed. After 10-15 minutes of explanation and practice with the apparatus, two consecutive experimental runs were carried out. The instrumentation and the routine were the same as for the normal subjects except that

1) Repeated blood pressure measurements were made by automated sphygmomanometry (Air-Shields), and the times of measurement marked on the UV trace.
2) No measurement was made of end-tidal CO_2 concentration (Fig. 5:24)

Extraction and treatment of data

(1) Values for ventilation ($\dot{V}_I$) and for heart rate (h) were obtained for the periods

i) Last 2 minutes of air-breathing

ii) Last (usually 3rd) min breathing 10% oxygen

iii) 10th to 60th sec after switch to 100% oxygen

(as for normal series except that '$H_{2,3}$' data was not

separately extracted). Heart rate in this series could usually be read directly from the calibrated trace, as there was less fluctuation than in normal subjects.

(2) The heart rate corresponding to each separate estimation of blood pressure was noted.

(3) As for normal subjects, the trace covering the period of reoxygenation was examined for a clear decrease in heart rate and/or ventilation, and values obtained for the elapsed time from the beginning of the first breath of oxygen.

Analyses

In this way 8 sets of data were assembled. For various technical reasons all were not complete for all measurements.

Heart rate: 6 complete sets of results were available for both postures and all three oxygen levels (air, last min hypoxia, oxygen). A 3-factor analysis of variance was carried out to assess the effect of oxygen level on heart rate. Further analyses were carried out for the two postures separately.

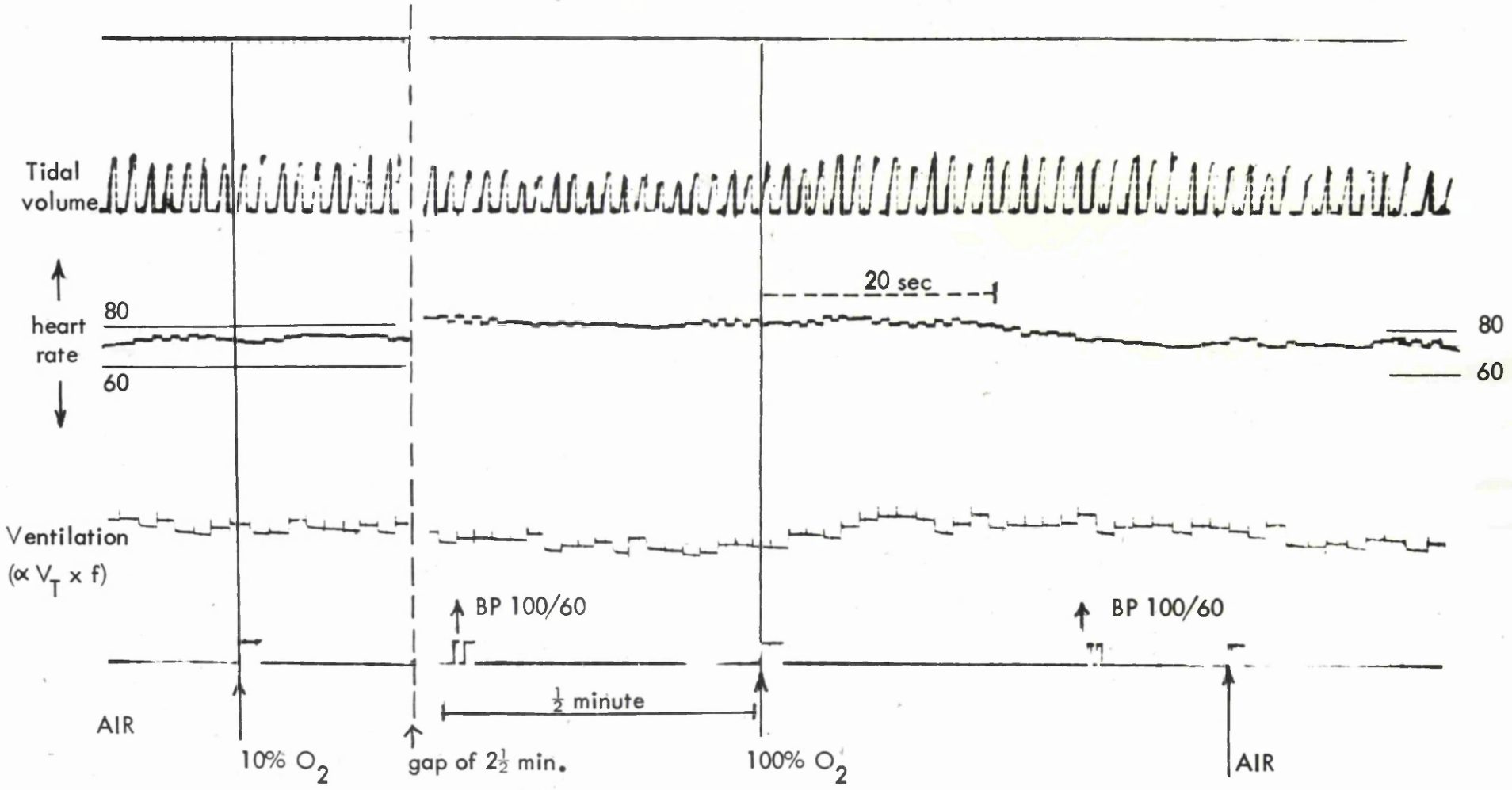
Since changes in the lying position were more consistent and greater in extent than those in the sitting position, and since the normal subjects were studied semi-recumbent, most other analyses were carried out on the results for the lying position only.

Ventilation: Complete replicated data for 5 subjects were analysed.

Blood pressure: Complete replicated data for 6 subjects were analysed, in terms of mean, systolic, and pulse pressures.

In addition to these analyses, the means, SD's and SE's for each variable were found, for all available runs, and for the changes in ventilation and in heart rate respectively as % reference value; comparisons were made by student's t-test between the values for air and hypoxia, and for hypoxia and oxygen.

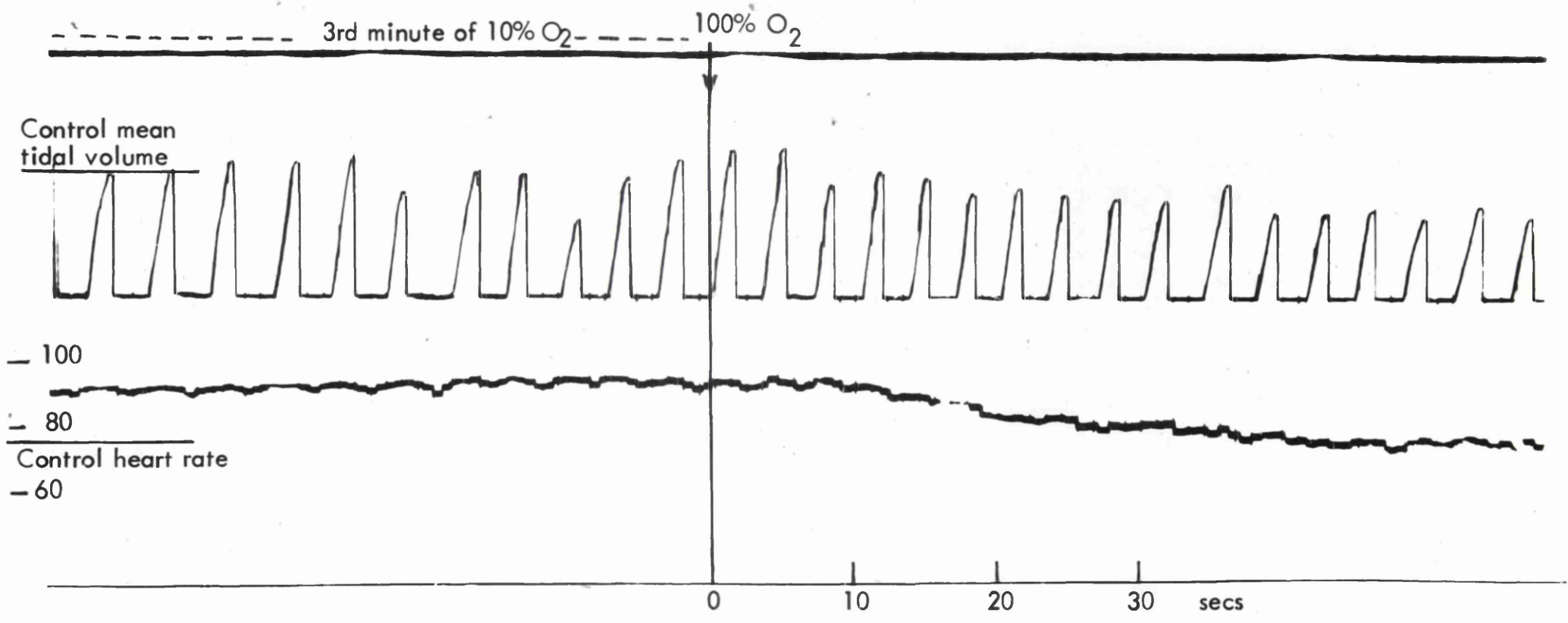
Fig 5:25a



Subject no 5 with cervical spinal cord transection (sitting)

Left : reference levels, breathing air; start of 10% oxygen.
Right: last half-minute on 10% oxygen, showing heart rate 10 b.p.m. higher than initial level; change to 100% oxygen shows decrease in heart rate after 20 secs, accompanied in this instance by an increase in ventilation.

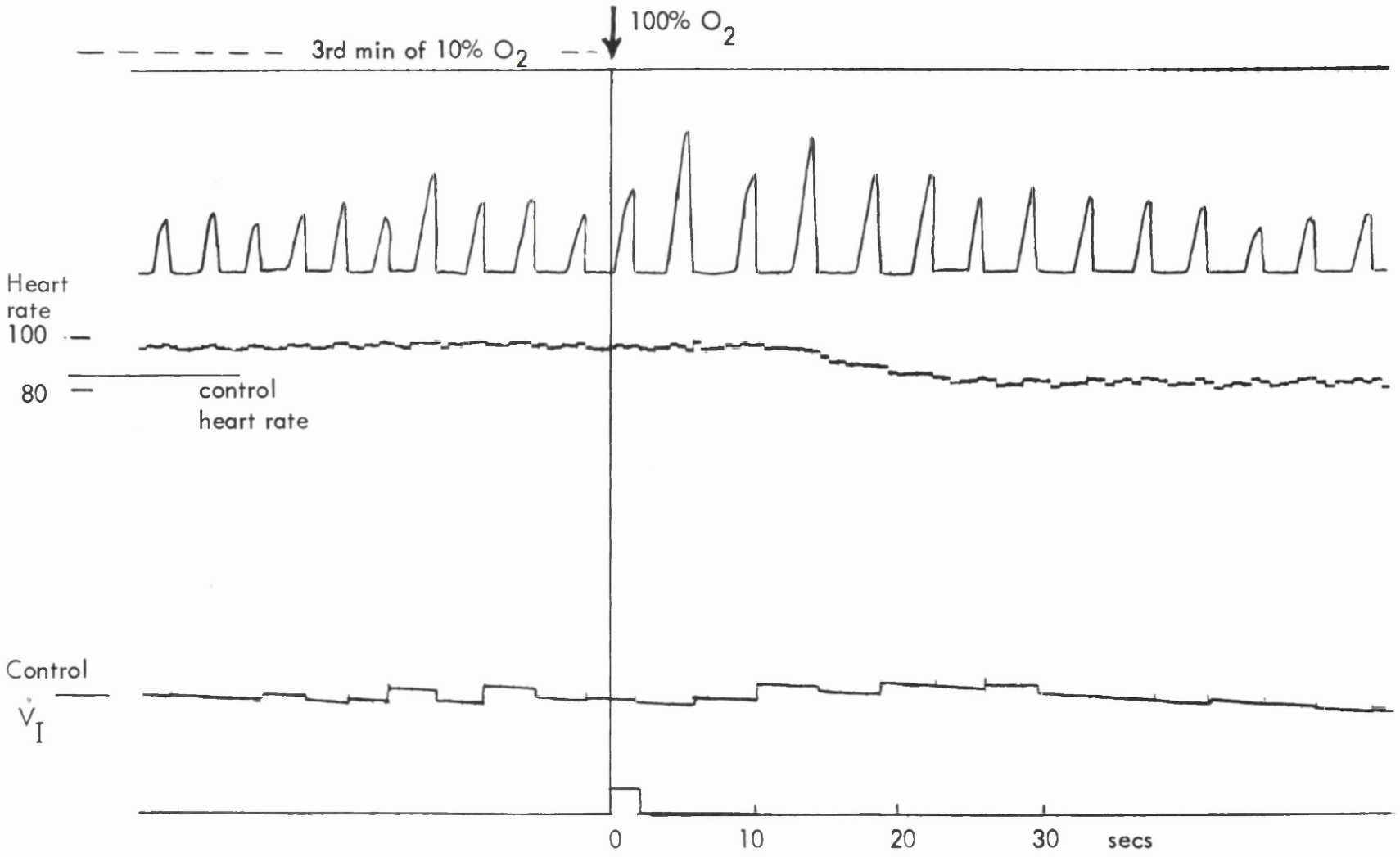
Fig 5:25b



Subject no. 4 with cervical spinal cord transection.
Increase in heart rate by 14 b.p.m. by the end of 3 min breathing 10% oxygen.
Decrease in heart rate starting 13 sec after switch to 100% oxygen.

Subject 7, lying

Fig 5:25c



Subject no. 7 with cervical spinal cord transection.

In this case the heart rate, but not the ventilation, increased during hypoxia. Heart rate decreased abruptly 15 sec after the switch to oxygen.

RESULTS

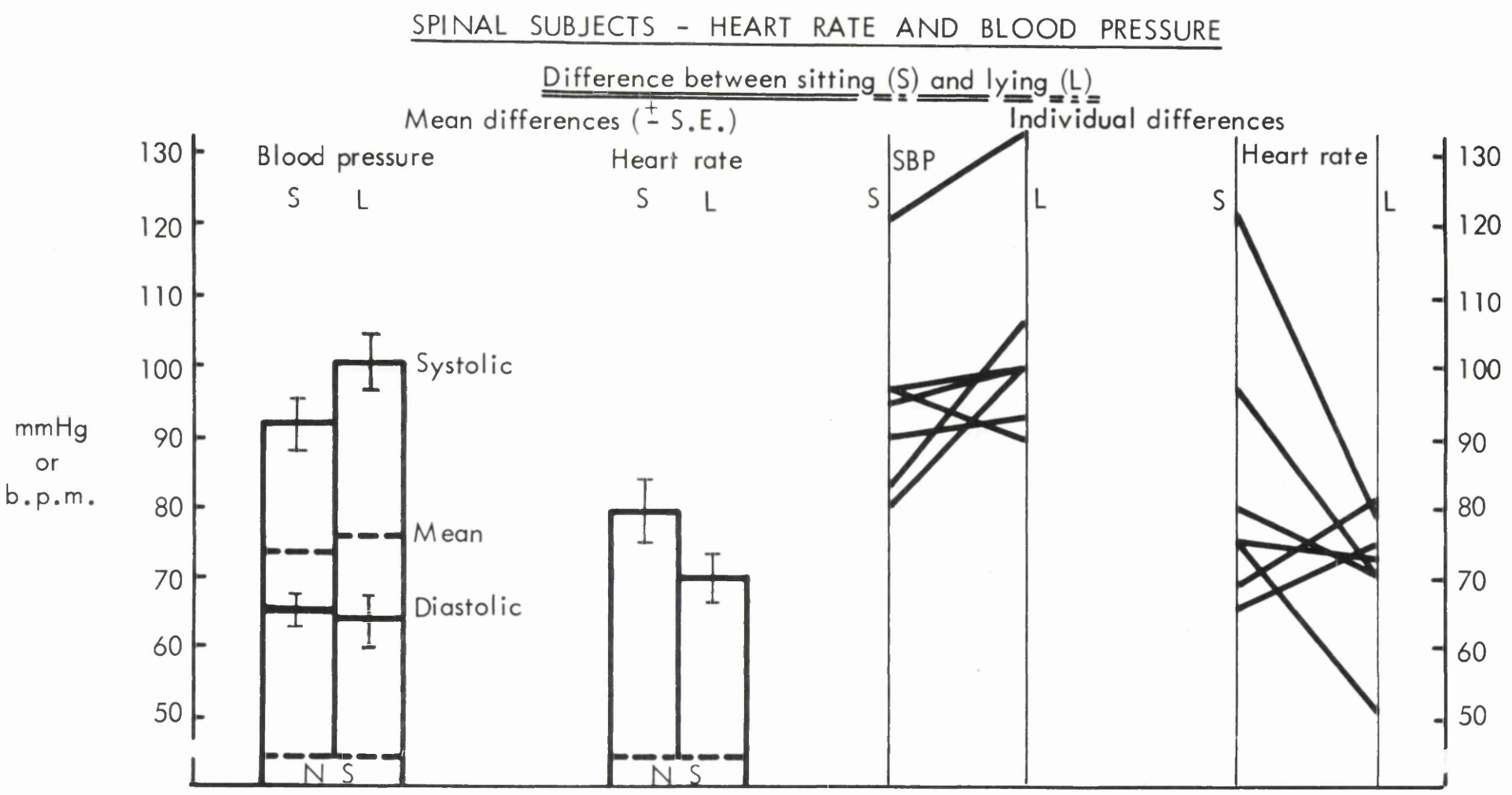
Representative records are shown in Fig. 5:25.a,b,c.

The heart rate increased in most instances during hypoxia, particularly when the subjects were studied lying. There was a decrease in heart rate at reoxygenation which was similar in magnitude and in timing to that recorded in normal subjects. Fig. 5:25c shows this decrease in heart rate, occurring within 20 sec of the first breath of oxygen from the hypoxic level of 97. b.p.m. to the initial airbreathing level of 86 b.p.m. (The heart rate scale on these traces is 50% smaller than for the normal series: the apparent change on a 'spinal' trace needs to be doubled to compare directly with a 'normal' trace).

The examples shown are notable for the absence of a decline of ventilation at reoxygenation: this was a consistent difference from the normal findings, and will be discussed (compare Fig. 5:20, p.5/67).

The results will be presented in detail; firstly, the cardiovascular changes: the differences in baseline values according to posture, analysis of the changes during hypoxia and reoxygenation in heart rate and in blood pressure, and of the relationship between the changes in these two; secondly, the ventilatory changes.

Fig 5:26


SPINAL SUBJECTS - HEART RATE AND BLOOD PRESSURE
Difference between sitting (S) and lying (L)
Mean differences (± S.E.)
Individual differences
Blood pressure
Heart rate
SBP
Heart rate
S
L
Systolic
Mean
Diastolic
N S
mmHg
or
b.p.m.
130
120
110
100
90
80
70
60
50

Heart rate and blood pressure in the two postures (Fig.5:26)

Breathing air

The values for heart rate and for blood pressure in the two postures respectively are shown in Fig. 5:26. On average the heart rate was lower and the blood pressure higher when lying, and this was more marked, as might be expected, in the more recently injured patients (Table 5:13).
Mean values for all measurements on all subjects, breathing air, were compared by t-test: the differences between the two postures were not statistically significant due to wide differences between subjects, and between the effect of posture in the different subjects.
These values do not refer to changes of posture on the same occasion, but to measurements made in the two different postures on separate occasions.

During air-hypoxia-oxygen sequence

The changes in heart rate which occurred were on average greater in the lying than in the sitting posture. Some analyses described below deal with 'lying' values only both for this reason and for comparison with the normal subjects who were studied semi-recumbent.

Changes in heart rate during hypoxia and reoxygenation

The mean values are shown in Fig. 5:27, together with the significance of comparisons between the air, hypoxic, and oxygen periods. The increase was by about 16%, or 10 beats per min lying, and both this and the decrease on reoxygenation were significant. (In the sitting posture, the average increase was 8%, 5 beats per min, and was not significant).

Fig 5:27

BLOOD PRESSURE AND HEART RATE BREATHING AIR (A), IN 3rd MIN OF 10% O_2 (H) AND 1st MIN OF OXYGEN (O)

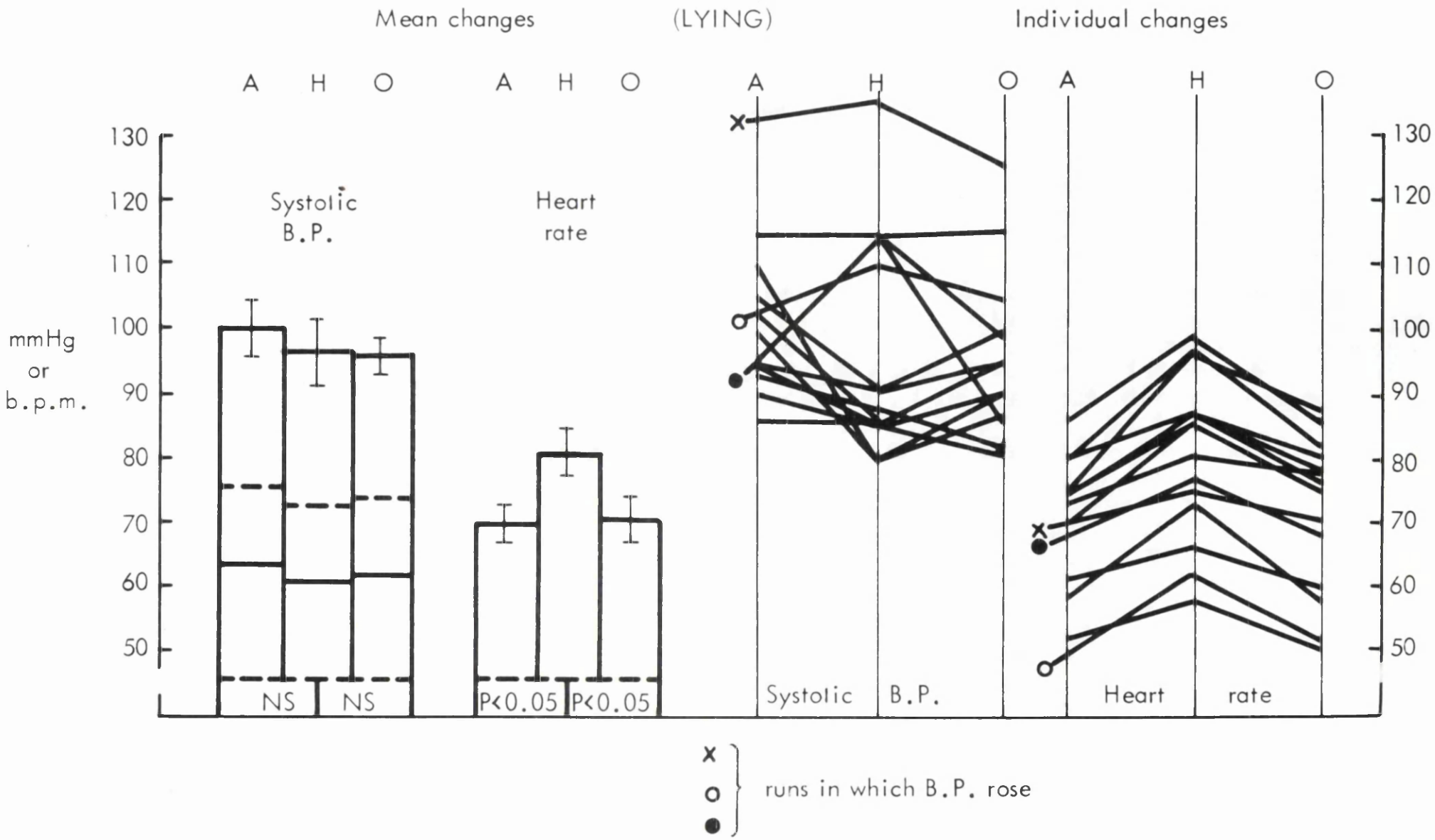


Table 5:14

Changes in heart rate

Mean results and comparisons between spinal and normal subjects by analysis of variance.

7 spinal, 7 normal subjects

(2 experiments on each)

	Air	10% O_2	
Spinal	70	82	N.S.
Normal	66	75	

P 0.05

4 spinal, 4 normal subjects

(2 experiments on each)

	Air	10% O_2	100% O_2	
Spinal	71	83	74	N.S.
Normal	69	80	68	

Extent of changes in heart rate

Further statistical analysis

The analysis of variance for 6 subjects led to the following conclusion:

The effect of O_2 level on heart rate was highly significant in either posture (lying, $p < 0.001$; sitting, $p < 0.01$).

Mean values for these subjects are shown in Table 5:15

Comparison with normal subjects (Table 5:14)

7 normal and 7 spinal subjects were compared by analysis of variance for the effect of hypoxia on heart rate. The effect of O_2 level on heart rate was again shown to be significant overall, and there was no variation in this effect attributable to spinal transection.
4 complete comparable replicated experiments for normal and spinal subjects respectively were analysed to compare both the change to hypoxia and the change to oxygen: this analysis also showed no significant variation in O_2 effect attributable to the spinal lesion.

Timing of the decrease in heart rate on reoxygenation

The first clear fall in heart rate (Fig. 5:25) could usually be defined readily, as in normal subjects and occurred at an average of 18.8 sec after the beginning of the first breath of oxygen.
This was suggestive of a longer delay time than in the normal subjects (mean 15.9 sec) but comparison (t-test) of the 16 spinal with 30 normal runs showed no significant difference.

Changes in blood pressure during hypoxia and reoxygenation

In most runs, in either posture the systolic blood pressure decreased during hypoxia (Fig. 5:27).

Table 5:15

Mean results for 56 subjects from analysis of variance

		Air	10% O_2	100% O_2
Heart rate b.p.m.	Sitting (6)	79 ⟶	84 ⟶	78
	Lying (6)	69 ⟶	81 ⟶	71
B.P. Systolic	Lying (6)	100	97	97
Mean mmHg		74	70	71
Ventilation L/min	Lying (5)	8.2 ⟶	9.1 ⟶	7.5

(⟶ : significant difference)

Although there was an average decrease, the three exceptions (Fig. 5:27) made this non-significant.

Extent of changes in blood pressure

Further statistical analysis

The analysis of variance for 6 subjects, lying, led to the following conclusion:

The blood pressure was very variable between subjects, and the effect of O_2 level on it was variable: no significant effect of O_2 level could be demonstrated, whether systolic, diastolic, or mean values were analysed.

The mean values for these subjects are shown in Table 5:15.

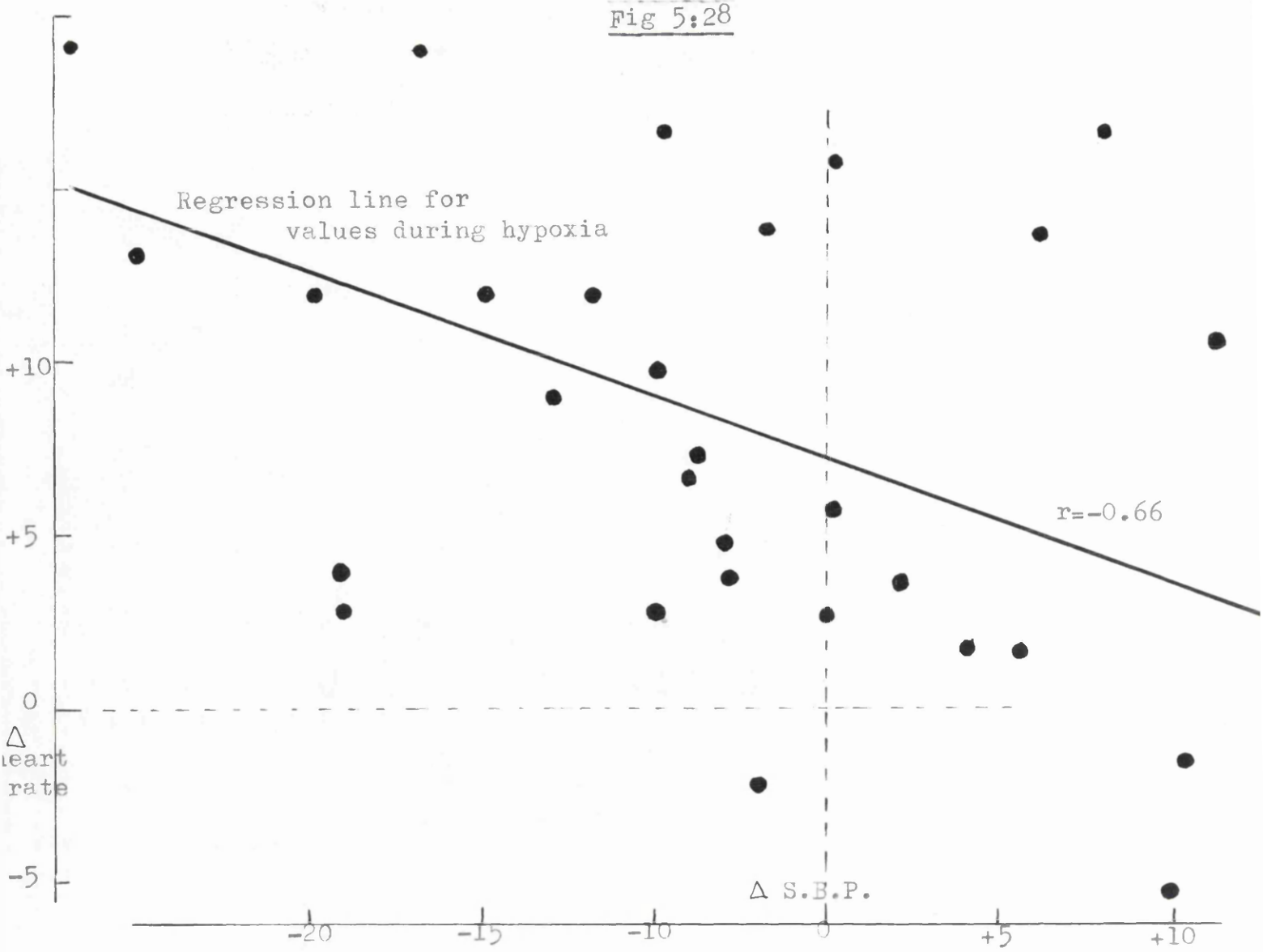
Relationship between changes in heart rate and changes in BP

Despite the non-significance of the blood pressure changes, it seemed possible that an effect of hypoxia in reducing blood pressure might simply be obscured by a few spurious increases in BP, which frequently occur in these patients. As such a fall in blood pressure might be held to account for the increase in heart rate, some care was given to further study of the relationship between individual blood pressure and heart rate changes.

i) All values obtained for BP during each run were matched with heart rate values from the continuous traces. This yielded a greater number of separate pairs of measurements than were used in the analysis for the standards measurement periods.

Blood pressure was plotted against heart rate, and correlation coefficients found, for each individual experiment. In 15 experiments analysed in this way, there were 4 in which the correlation was significant

Fig 5:28



Deviations from mean reference air-breathing value of systolic blood pressure and of heart rate. 8 spinal subjects, lying. Each point represents a single pai observations during hypoxia.

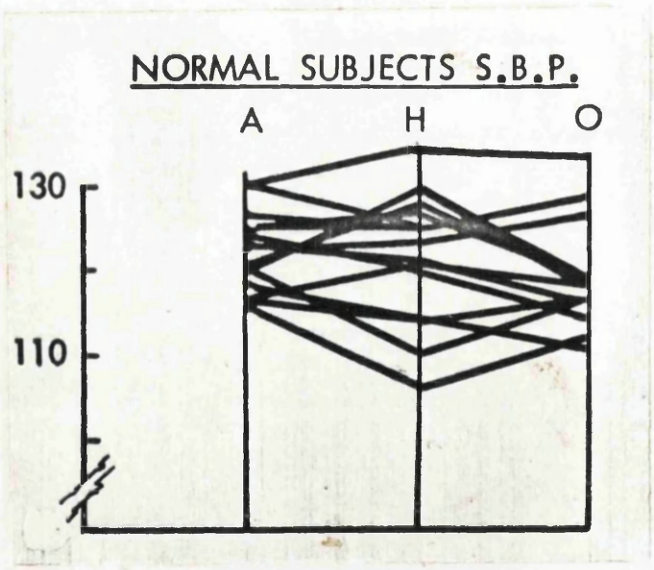


Fig 5:29

Systolic blood pressure changes in normal subjects

A, reference , air-breathing
H, last min of hypoxia
O, first min of oxygen

Compare Fig 5:27 for spinal subjects.

(subjects 2 and 5 sitting, subjects 3 and 6 lying). There was no indication that the points for hypoxia might lie on a different line from those for air and oxygen (see Discussion, p. 5/91)

ii) In order to pool the results for all subjects, each individual BP and heart rate value was expressed as a difference from the subject's reference value. Change in BP was plotted against change in heart rate. There was considerable scatter, but the correlation was significant (r -0.31; $p<0.01$). Separate correlations were calculated for (i) values during hypoxia and (ii) values during air and oxygen (Fig. 5:28). The changes during hypoxia only, showed a close relationship (r -0.66, $p<0.001$), and the others no correlation at all.

The significance of this is doubtful although it suggests a possible cause-effect relationship during hypoxia. On this account additional runs were carried out on normal subjects in order to take comparably frequent blood pressures and be sure of the absence of such a relationship.

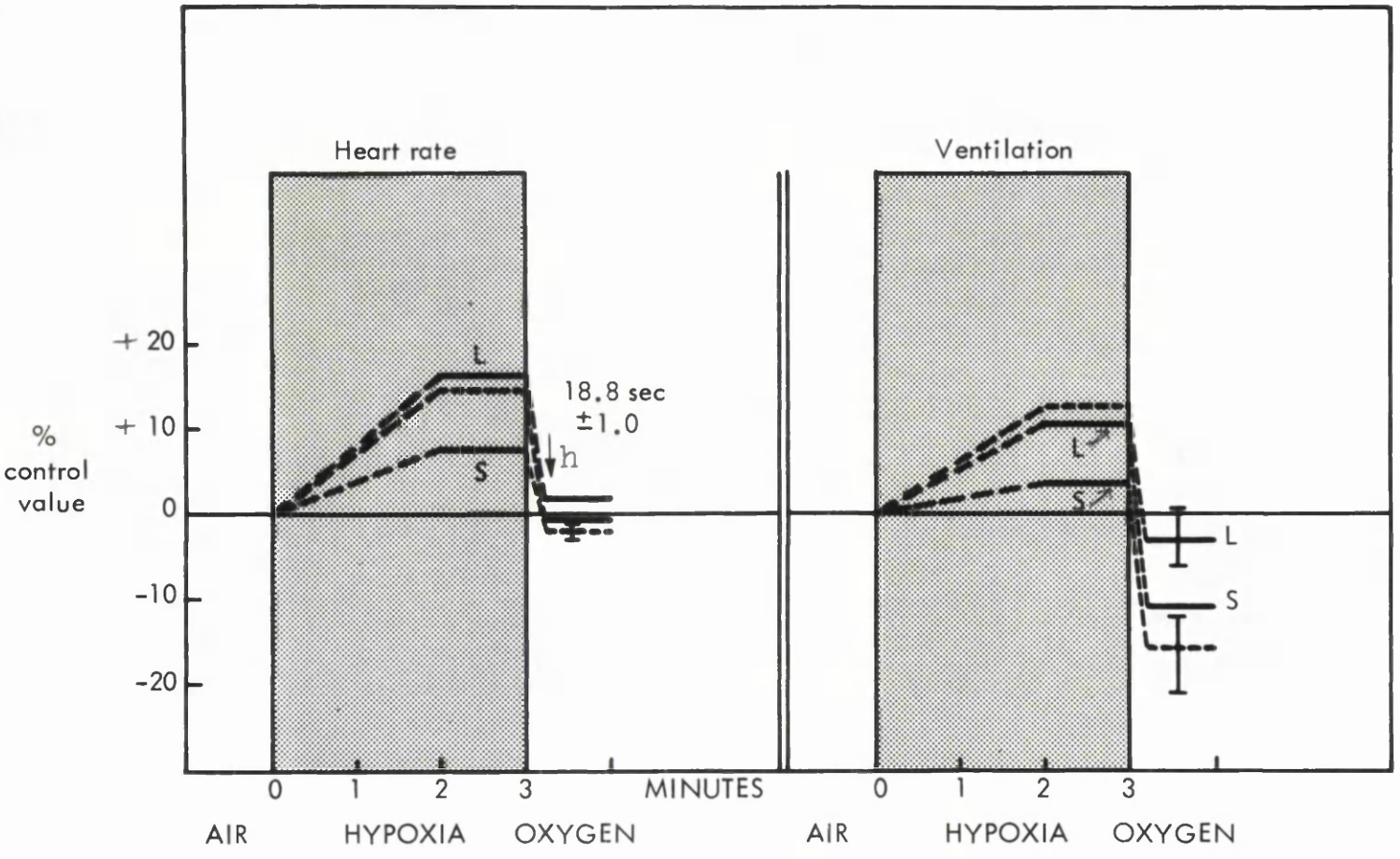
These results combined with occasional earlier observations are shown in Fig. 5:29. Clearly there is no consistent fall of BP in normal subjects during hypoxia.

VENTILATION

Ventilation increased in almost all instances during hypoxia, and by an average of 11.5% (lying) (Fig. 5:30). The reduction of ventilation at

Fig 5:30

CHANGES IN HEART RATE AND VENTILATION DURING HYPOXIA AND AT REOXYGENATION



Comparison of 'spinal' with normal subjects

L } 'spinal' } lying
S } patients } sitting
--- Normal subjects (30 runs in 5 subjects)

Mean results from 14 runs on 8 spinal subjects, in each position.

↓h : interval before decrease in heart rate in spinal subjects
Bars: S.E.M. reduction of ventilation at reoxygenation.

reoxygenation was less than in normal subjects, though the difference between the mean changes from hypoxia to oxygen did not reach significance (note large standard errors, Fig. 5:30).

A. Extent of changes

Statistical analysis

(1) The conclusions from an analysis of variance for five subjects with complete data in the lying position were:

The effect of oxygen level on ventilation was highly significant ($p < 0.001$) and there was no significant difference between its effects from subject to subject, although the actual difference in baseline ventilation between subjects was considerable.

The mean values for these 5 subjects are shown in Table 5:15.

(2) Comparison between mean percentage changes for all runs

This method of analysis showed no significant changes from air to hypoxia, or from hypoxia to oxygen,

on account of the variation between subjects: this demonstrates the value of the analysis of variance.

B. Timing of decrease of ventilation on reoxygenation

It was virtually never possible to define a clear decrease in ventilation after the first breaths of oxygen.

DISCUSSION

The methods of recording, and validity of findings were as already discussed for heart rate and ventilation.

An approximate calibration for the arbitrary units of $\dot{V}_I$ was obtained by measuring expired volume from time to time in some subjects with a Wright anemometer and comparing this with the continuous trace. This gave

some check, in the absence of CO_2 recording, on the normality of the level of ventilation. The mean resting airbreathing ventilation was 8.2 L/min by this calculation, which was comparable with the early normal series, indicating that there was unlikely to have been any serious discrepancy between the spinal and normal series in terms of hyper- or hypo-ventilation.

The measurement of blood pressure was not always completely straightforward and reliable. The placing of the stethoscope diaphragm on a flexed elbow, and of the pulse monitor on a flexed finger presented some problems, and there were occasional artefacts and failures.

Significance of Results

Ventilation

It was anticipated, and observed, that the ventilatory response to hypoxia was still present after cervical spinal transection. It was as normal despite intercostal paralysis. However, the usual ventilatory depression at reoxygenation was lacking, although the difference from normal could not be shown to be statistically significant by taking and comparing the

mean values over the selected periods (Fig. 5:30) Further study would be necessary to confirm the difference, and it would have to be related confidently to similar ventilation in terms of PCO_2. If this is a real difference, however, it seems unlikely that absolute ventilation level would account for it: there was no suggestion in either the spinal or the normal series that the decrease on reoxygenation was correlated either with the initial ventilation, or with the increase during hypoxia. It could be tentatively suggested that a difference might occur because of absence of intact sympathetic efferent pathway from medulla to carotid bodies. The autonomic control of carotid body blood flow and oxygen consumption has been shown to be of some significance in the determination of sensitivity to hypoxia. Since the decrease of ventilation on cancellation of the hypoxic drive probably reflects the afferent chemoreceptor traffic during hypoxia more accurately than does the actual ventilatory response, then estimation of this decrease could be a valid method of investigating the effect of the lack of sympathetic control of chemoreceptors in man.

Cardiovascular changes

Responses to hypoxia

The response of heart rate was similar in both timing and extent to that in normal subjects; this suggests, but does not prove, that the normal mechanism is intact.

Heart rate response in relation to blood pressure changes; a baroreceptor heart rate reflex must be considered as a cause of tachycardia in the spinal subjects: there was a suggestion of an inverse relationship between the two, which might have been more significant if blood pressure measurements had been more frequent and reliable. Heart rate in such subjects rises in response to fall of blood pressure, since the baroreceptors and vagi are intact. From the data of Johnson, Crampton Smith & Spalding (1969) for a similar patient during repeated postural changes, a blood pressure-heart rate response line can be drawn and is of similar slope to the regression line found in the present series during hypoxia.

A fall of blood pressure during hypoxia is theoretically more likely in these subjects than in the

normal: the direct effect of low PO_2 in reducing peripheral resistance could be counteracted neither by reflex vasoconstriction via the baroreceptors, nor by a direct effect of hypoxia on the vasomotor centre, since both depend on the brain-spinal cord sympathetic pathway.

However, for BP changes to be put forward as the cause of tachycardia, the BP would have to be shown to be reversed almost immediately on reoxygenation. This did not appear to happen (Fig. 5:28).

Hypoxia of the isolated cord might, however, increase the sympathetic outflow: this may account for the reduction in blood pressure not being greater than it was. This possibility must also be considered as a mechanism for increasing cardio-accelerator activity (Alexander, 1945).

To settle this point, it would be necessary to plot a BP - heart rate (or pulse interval) curve in the manner of Pickering, Sleight & Smyth (1968), for any given patient, and compare the relationship during hypoxia with that in normoxic conditions: if these subjects behave like normal subjects, and have some

other cause for their tachycardia than the BP changes, the hypoxic response line would be parallel to the normoxic curve, but shifted towards a higher heart rate (Bristow, Brown, Cunningham, Goode, Howson & Sleight, 1958).

Responses at reoxygenation

Relationship between heart rate and ventilation

The results for spinal patients confirm the dissociation between the two responses which was observed in some normal subjects and in the maintained ventilation experiments. A clear fall in heart rate was readily definable in the spinal subjects, but a clear fall in ventilation was not.

Timing of heart rate change

On average the time delay between the first breath of oxygen and the decrease in heart rate was slightly longer in the spinal subjects than in the normals; this did not however reach statistical significance. The range of delay times was smaller - there were no instances of a fall in heart rate occurring in less than 14 sec in all 30 runs on the spinal subjects, whereas a reduction at about 10 sec occurred many times in the normal subjects. The instrumental dead space

was identical in the two series, and it is unlikely that differences in ventilation would account for any difference in delay time.

If the delay time in normal subjects were shown to be compatible with the time taken for oxygenated blood to affect the medullary centres, then it would be relevant to consider whether the delay time in spinal subjects is compatible with the time taken for oxygenated blood to affect the spinal centres. The paper of Alexander (1945) indicates a delay of 20 sec (allowing for dead space) between the first breath of pure nitrogen and the increase of sympathetic outflow activity in spinal cats; the precise timing of the response to reoxygenation is unfortunately not given, but would appear to be similar. The comparable delay time in the spinal patients was 10-15 sec (see p. 5/73) Reoxygenation of the spinal centres would appear to be a fairly unlikely explanation on these grounds.

There was never a hint of transient increase in heart rate on reoxygenation in normal and spinal subjects: theoretically, if the chemoreceptors primarily cause bradycardia, and if there was no accompanying change

in ventilation, and a longer pathway to the tachycardia-response receptor site than to the chemoreceptors, a transient primary chemoreceptor effect might have been anticipated.

THE EFFECT OF HYPOXIA AND REOXYGENATION ON A PATIENT FOLLOWING BILATERAL REMOVAL OF CAROTID BODY TUMOURS

McG., aged 48 was first studied (by permission of Professor Bryan Jennett) 3 weeks after the removal of a carotid body tumour from the right side of the neck; a similar tumour had been removed from the left side a year before. He was studied a second time 2 months later.

METHODS

First occasion:

Hypoxia-reoxygenation experiment: the routine sequence previously applied to normal and spinal subjects was used; CO_2, heart rate and ventilation were recorded. Two complete runs were carried out.

Valsalva manoeuvre: the subject held his breath with a positive pressure of 35-40 mm Hg for 10 sec; the effect on heart rate was recorded. This was repeated twice.

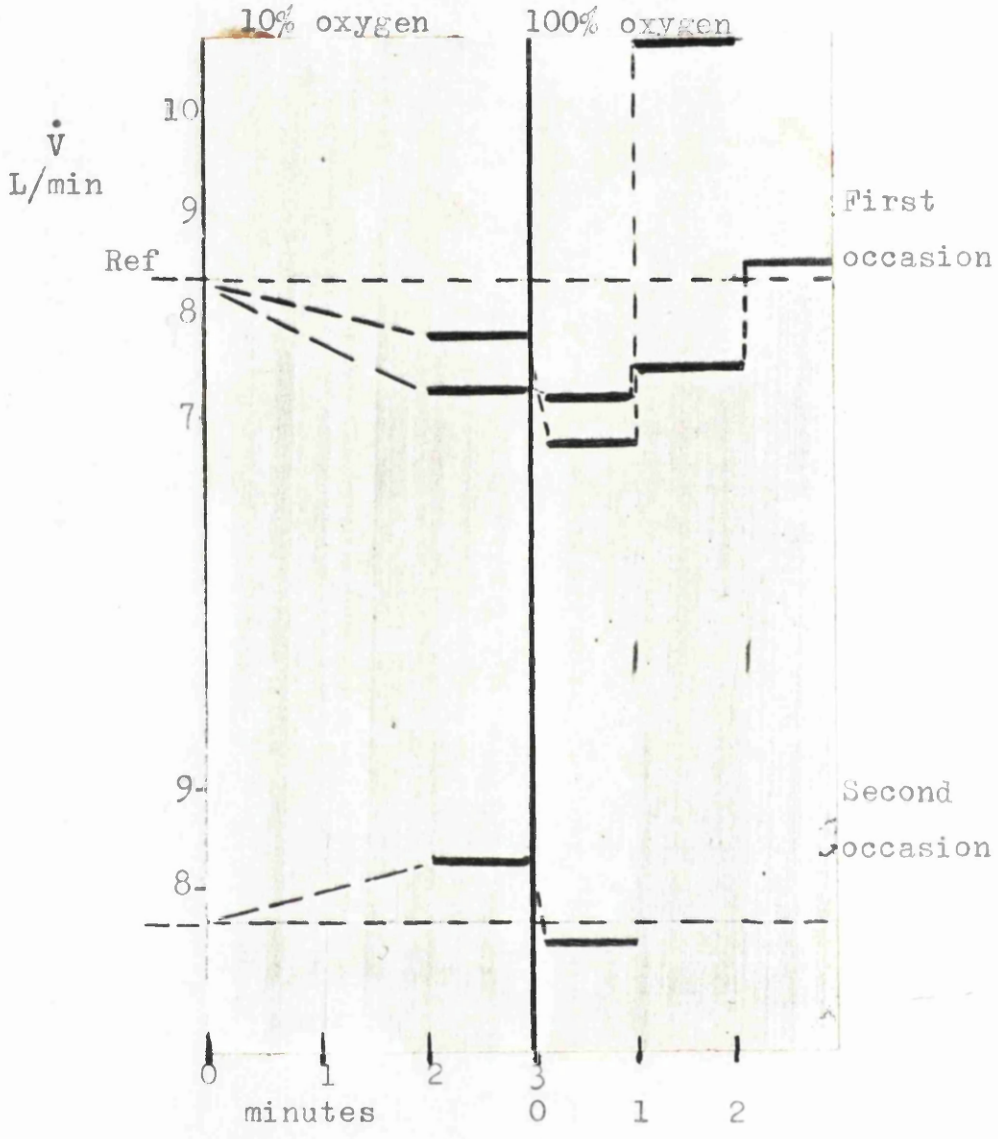
Second occasion:

The procedures of the first occasion were repeated. In addition:

CO_2 response tests in high and in low oxygen were carried out by a re-breathing method. The test in high oxygen was similar to that employed previously by us in normal subjects,

Fig 5:31

McG. Response of ventilation during hypoxia and reoxygenation following bilateral removal of carotid body tumours.



(Jennett, 1968); a 4 litre bag of 7% CO_2 in oxygen is used; the subject rebreathes from this for 3-4 min, during which time the CO_2 concentration rises linearly (Read & Leigh, 1967); ventilation and CO_2 concentration are continuously recorded, and from the records a response curve of ventilation against alveolar PCO_2 is plotted, using values calculated for each half minute. A similar test starting with 7% CO_2 in air was also applied; a gas sample was circulated through the oxygen analyser continuously to warn against a severe degree of hypoxia.

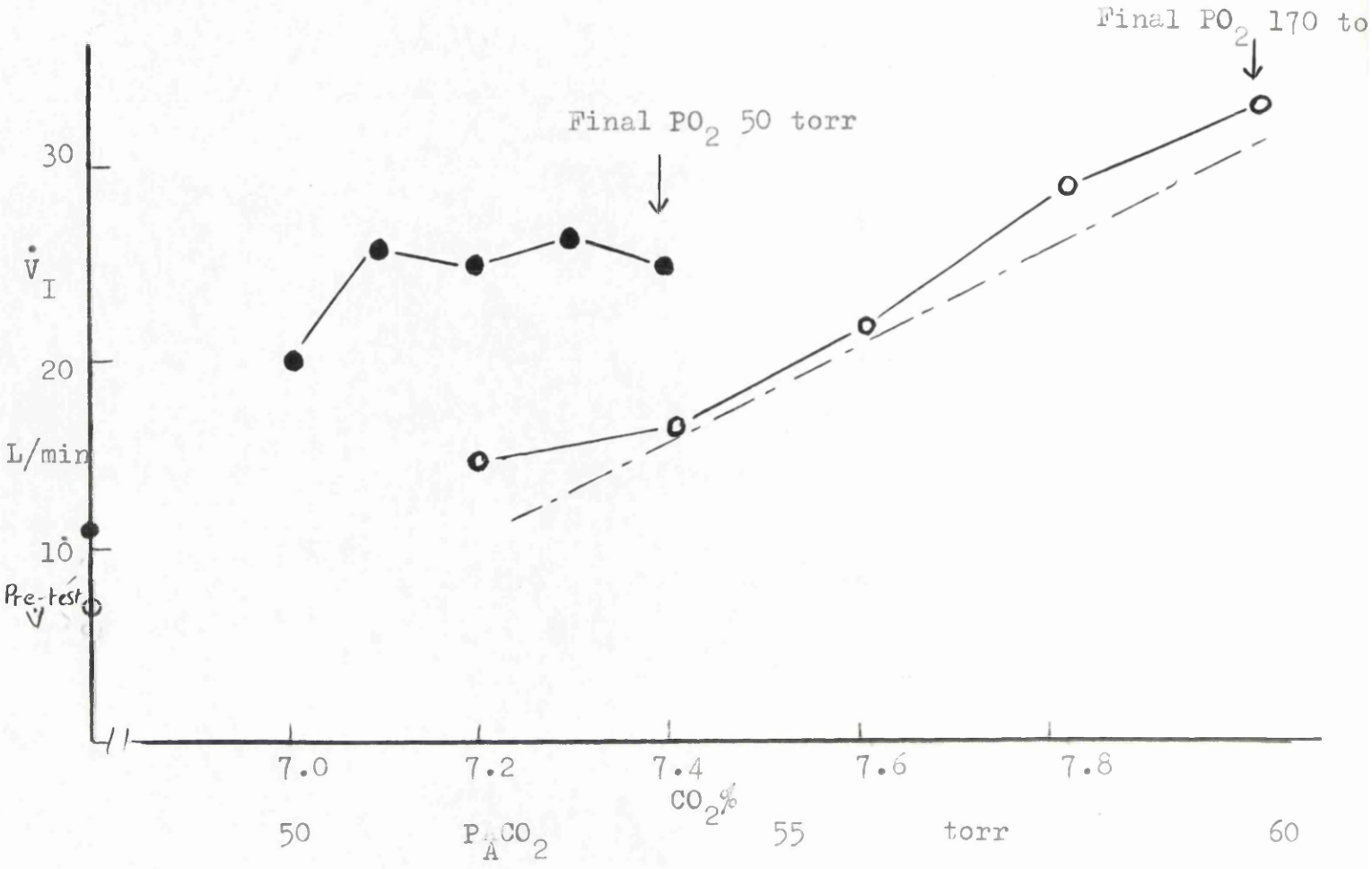
The effect of tilting on heart rate was recorded, employing abrupt changes from reclining-seated, to a position with the head low and feet raised.

RESULTS

VENTILATORY RESPONSES

The resting air-breathing PCO_2 was 38-39 torr; the patient may, however, have been over-breathing compared to his normal post-operative state

Fig 5:32



Open circles : Rebreathing CO_2 in oxygen

Solid circles : Rebreathing CO_2 in air

--- . - . - . - Mean slope and position of normal response line.

in the unfamiliar laboratory surroundings).

Hypoxia-reoxygenation

Fig. 5:31 shows the measurements made from the traces treated in the same way as for the normal and spinal series (compare Fig. 5:30). On the first occasion ventilation decreased slightly during hypoxia in both runs; such a decrease could not be demonstrated to be significant from only these two estimations on a first occasion. There was, however, a further decrease in ventilation on reoxygenation, suggesting the presence of a hypoxic drive. Subsequently, after the first minute, ventilation increased.

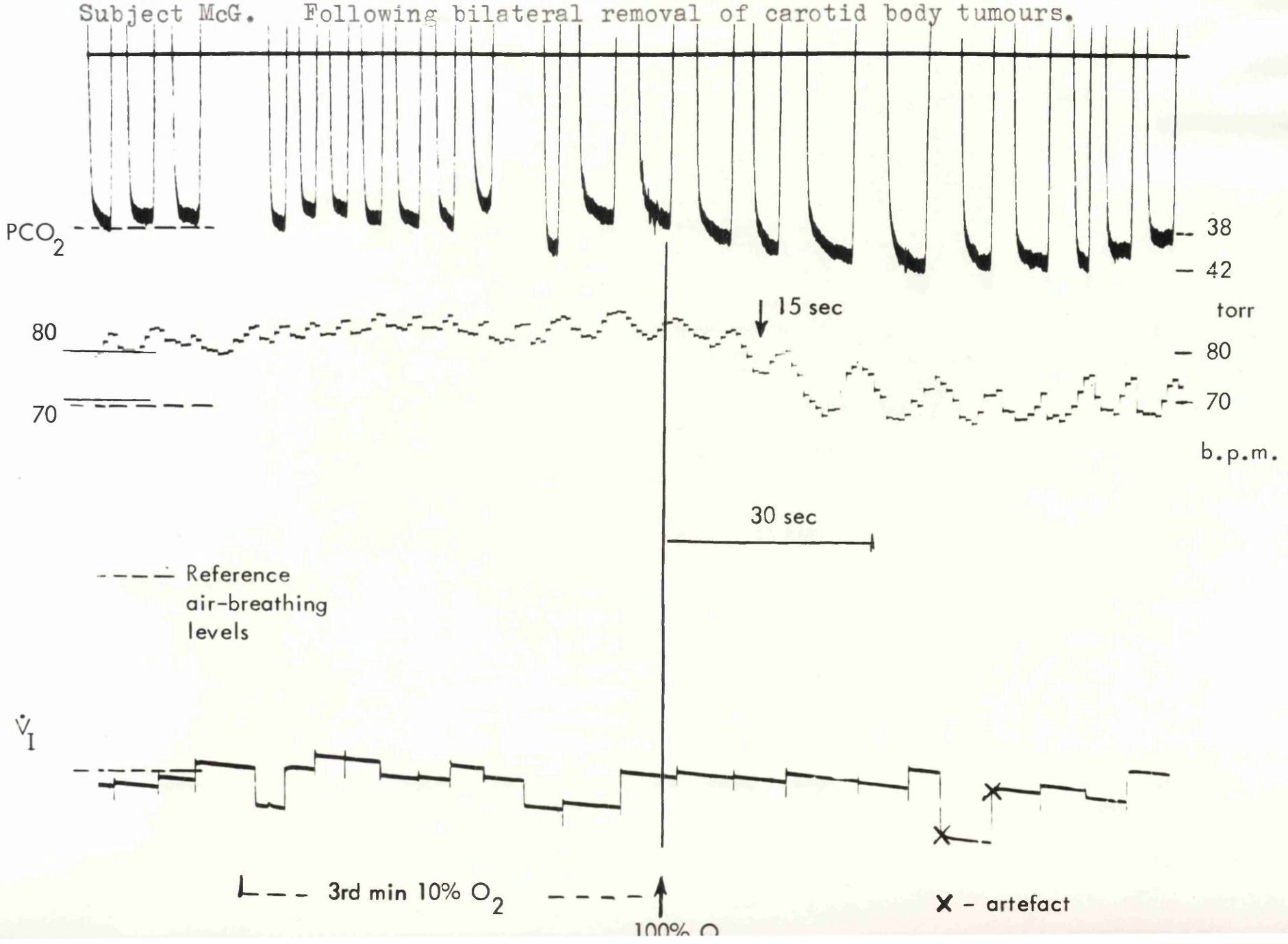
On the second occasion there was a small increase during hypoxia, and a decrease at reoxygenation.

CO_2 response tests (Fig. 5:32)

In oxygen: There was a linear rise of ventilation with increasing CO_2 concentration and the slope and position of the response line were very close to the mean for normal subjects.

In air: The test was terminated when the oxygen level in the bag reached 50 torr; there was no undue discomfort from hyperpnoea at this time. Although there was a steep increase in ventilation initially (perhaps due to psychological factors), the increase levelled out.

Fig 5:33


Subject McG. Following bilateral removal of carotid body tumours.
PCO2
38
42
torr
80
70
15 sec
80
70
b.p.m.
30 sec
Reference
air-breathing
levels
V̇I
3rd min 10% O2
100% O
X - artefact

Unfortunately it was not possible to repeat the test on this occasion.

HEART RATE RESPONSES

Hypoxia-reoxygenation

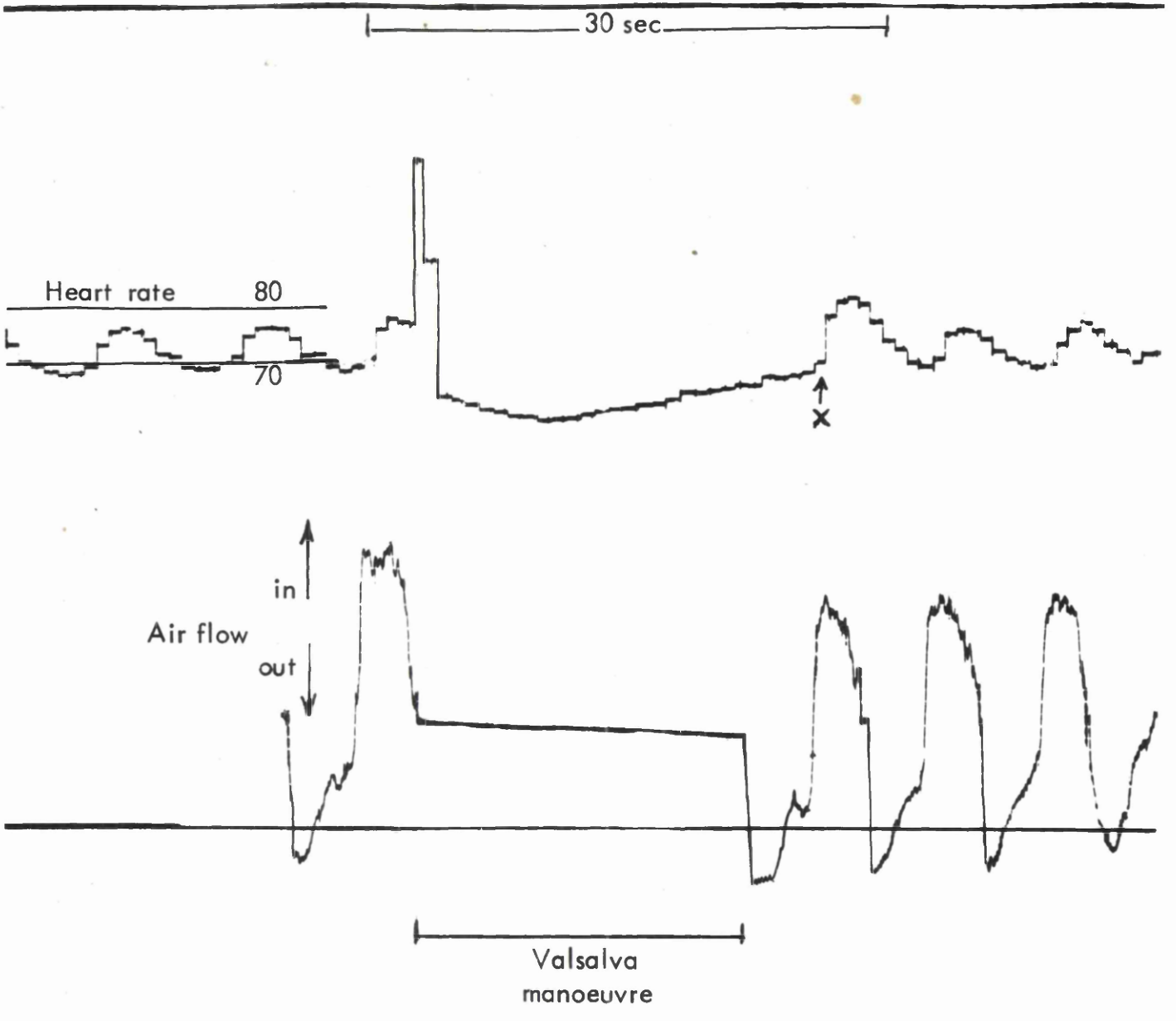
The response of heart rate to hypoxia and reoxygenation was similar to that in normal subjects, both in extent and in timing. Fig. 5:32 shows a run in which the abrupt decrease in heart rate is one of the clearest examples in the whole series. The result was similar on both occasions.

Valsalva manoeuvre

Normally a marked bradycardia accompanies the blood pressure "overshoot" about 4 sec after the release of intrathoracic pressure. In this subject there was no bradycardia: the heart rate simply increased as he took his first inspiration. This result was constant in three repeats on each occasion (Fig. 5:34). Tilting did not produce any definable change in heart rate.

Sinus arrhythmia was clearly defined throughout the recording (Fig. 5:33).

Fig 5:34



Subject McG : Following bilateral removal of carotid body tumours.

At X there would normally be a bradycardia.

DISCUSSION

It is unlikely that the nerve supply to the carotid sinus region would have survived the operations, even if any normal glomus tissue remained apart from the tumours. Destruction of the carotid baroreceptor and chemoreceptor activity could therefore be anticipated.

The reduction of ventilation, rather than an increase, during 10% oxygen breathing would not have been sufficient on its own to be taken as evidence of absent carotid chemoreflex function: there were occasional runs in normal subjects - first runs, on first occasions - when ventilation did not rise, or even decreased slightly. However, this was supportive evidence of chemoreceptor destruction. The indication of a remaining hypoxic drive, could be attributed to aortic chemoreflex activity: insufficient to counteract medullary depression and increase ventilation during hypoxia, but sufficient to account for the decreased ventilation on reoxygenation.

The result of rebreathing CO_2 in air is also suggestive of a very low responsiveness to hypoxia.

If, then, the carotid bodies were not active, it is confirmed by these results that the normal heart rate response does not in any way depend on them. The rise in heart rate was slightly above normal average, and the decrease at reoxygenation very brisk: it is possible that a greater tachycardia could occur in the absence of a normal chemoreflex vasoconstriction and attendent

tendency towards hypertension and baroreflex bradycardia.

The presence of such a marked sinus arrhythmia is of some interest: firstly, on account of the age of the subject - it was at least as marked as that found in several twenty-year-olds; secondly because it casts doubt on explanations for sinus arrhythmia which depend on an active baroreceptor-heart rate reflex (Davies & Neilson, 1967), unless this can be mediated entirely by the aortic receptors. The aortic baroreceptors were presumably intact in this subject, as well as aortic chemoreceptors: it seems surprising therefore that there should not be a heart rate response to tilting.

Holton & Wood (1965) reported no response to tilting for a few weeks after operation in their 2 subjects following carotid glomectomy: they did not observe the effect of Valsalva manoeuvre until 6 weeks afterwards, on account of the patients inability to perform it, and at that later time, there was a normal overshoot of blood pressure with a bradycardia. McG. was examined at a comparable time, on the second occasion: there was no sign of baroreceptor recovery, although there was of chemo-receptor recovery.

Conclusion

Observations on this subject add to the small amount of direct evidence in man that carotid chemoreceptors have a similar function to that in animals, in being necessary for the full ventilatory response to hypoxia. They also add to the less direct evidence already presented in this thesis that the changes in heart rate during hypoxia and reoxygenation are not associated either primarily or secondarily with a carotid chemoreceptor effect.

POSTSCRIPT

Experiment including intra-arterial blood pressure recording

By the cooperation of colleagues at the Glasgow Royal Infirmary and of subject "W" who volunteered, it was possible near the time of completing this thesis to carry out the hypoxia-reoxygenation sequence with continuous intra-arterial blood pressure recording (Devices strain gauge); intermittent arterial blood gas estimations were also carried out (Radiometer pH, PO_2 and PCO_2 electrodes).

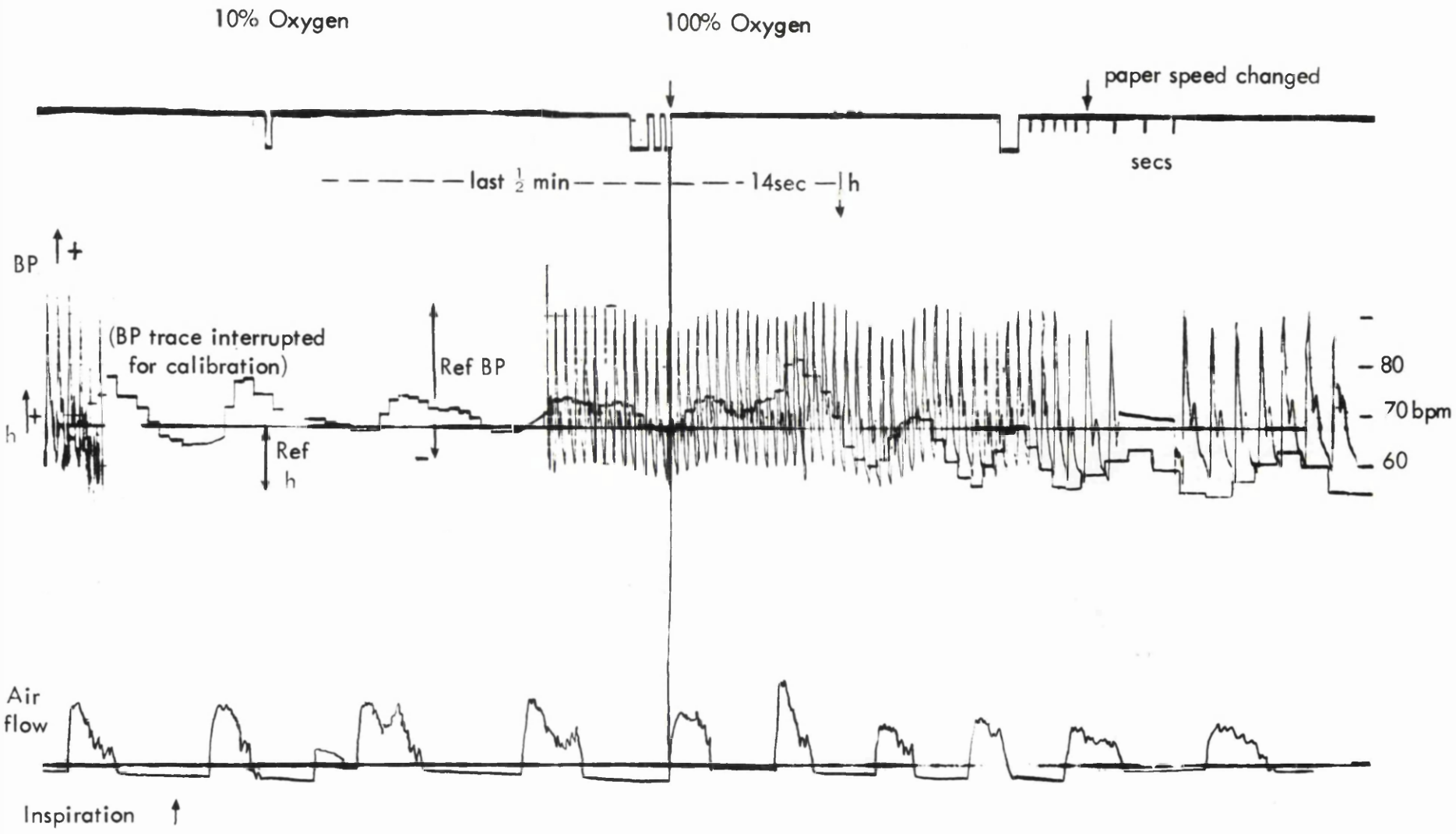
The intention also to record pulmonary artery pressures on this occasion was defeated by technical difficulties.

The procedure was essentially as in earlier experiments: three complete runs were carried out with an air-breathing reference period, 3 min 10% oxygen in one run and 5 min in the third, ½ min 100% oxygen. Heart rate, blood pressure and air flow were recorded.

RESULTS

Heart rate and BP

Despite the situation, the subject showed heart rate changes which were similar to his own mean results


10% Oxygen
100% Oxygen
paper speed changed
secs
last ½ min
14sec
h
BP
+
(BP trace interrupted for calibration)
Ref BP
Ref h
h
+
80
70 bpm
60
Air flow
Inspiration

in the earlier series of experiments. An increase by 6-10 beats per min was not accompanied by any change of blood pressure during the development of hypoxia; the actual level of systolic BP was, however, above this subject's normal, remaining steadily around 150-155 (normally 115-120). A decrease in heart rate occurred as usual shortly after reoxygenation (Fig. 5:35), and was quite definitely not secondary to even a transient or minimal rise in systolic or pulse pressure.

These findings were consistent in all three runs.

Arterial blood gas measurements

These confirmed that the degree of hypoxia, and the extent of the ventilatory response, were as found on average in the earlier series:

	P_aO_2	P_aCO_2	pH
AIR	80.7 torr	39.8 torr	7.410
HYPOXIA			
Run 1, at 3 min	42.0	36.8	7.436
Run 2, at 5 min	36.8	37.3	7.446
"OXYGEN"* Run 3, 1 min after first breath 100% O_2, after 3 min 10% O_2	99.7	41.6	7.397

* On air after ½ min 100% O_2.

Discussion

These results confirm the lack of any consistent change in blood pressure during this degree of hypoxaemia; they rule out any change in blood pressure as a cause of either the increase in heart rate during hypoxia, or the decrease at reoxygenation, in this normal subject.

The blood gas measurements are satisfactory confirmation of degree of hypoxaemia achieved, and of the end-tidal estimations made, by the techniques which have been applied in all earlier experiments.

GENERAL SUMMARY AND CONCLUSIONS FROM EXPERIMENTS ON RESPONSES TO HYPOXIA AND REOXYGENATION

Summary of findings

An increase in heart rate virtually always occurred during hypoxia induced by breathing 10% oxygen for 3-4 min. When hypoxia was continued for up to 10 min there was usually very little further increase.

The increase was independent of the initial heart rate, and occurred similarly in normal subjects, subjects with transection of the cervical spinal cord, and a subject with loss of ventilatory response to hypoxia.

The heart rate decreased to its initial value on reoxygenation, and this decrease was:

1) a constant finding, in all normal and abnormal subjects

2) usually abrupt, and after a delay time comparable to that before the fall in ventilation

3) not linked with the fall in ventilation: sometimes preceding it, often coincident with it, occurring also in its absence; and therefore not attributable to the fall in ventilation.

Relevance of these results to possible mechanisms for hypoxic tachycardia

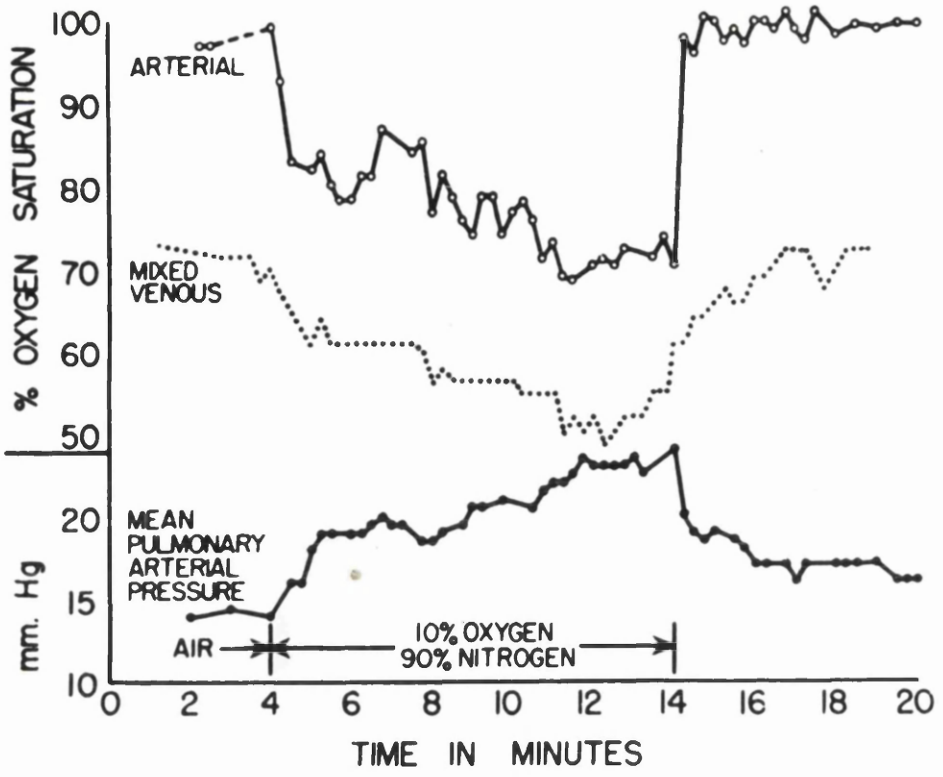
a) Receptor sites

The timing of heart rate response at reoxygenation would have to be compatible with the time taken for oxygenated blood to reach the suggested site. Possibilities are:

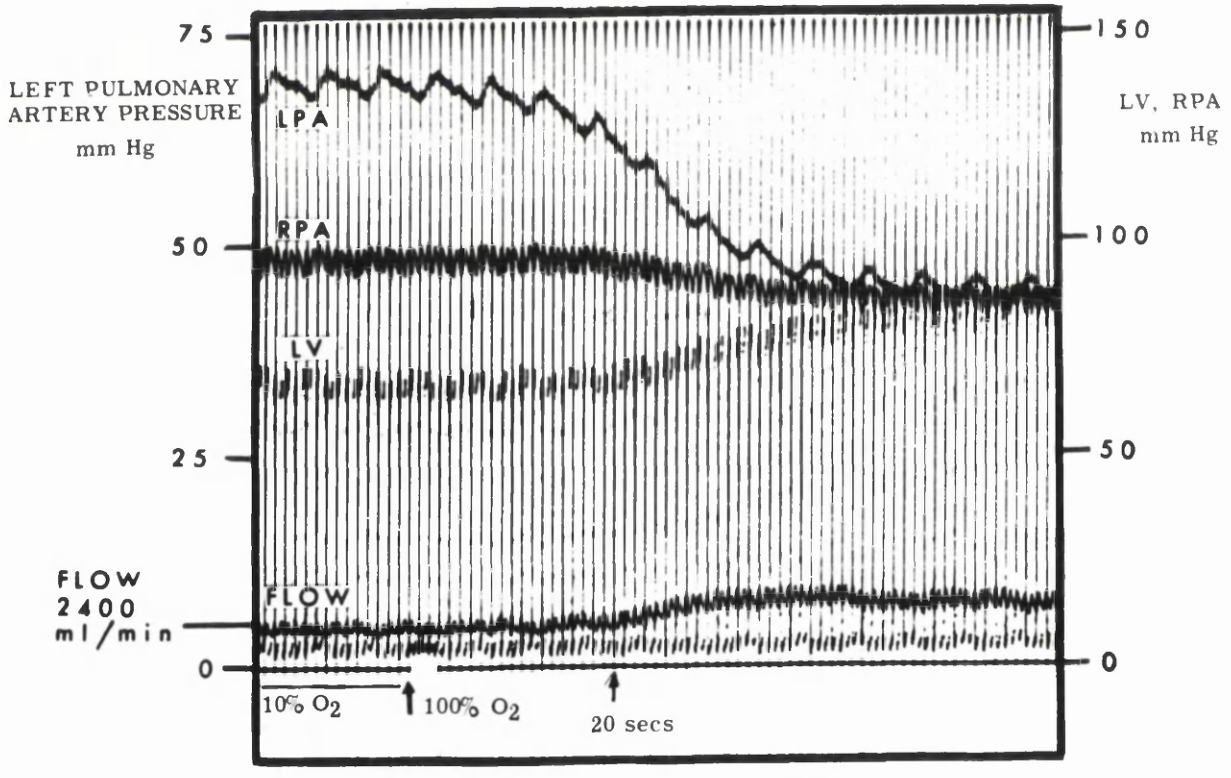
i) arterial chemoreceptors: the carotid bodies cannot be the receptor site since the response is normal after their removal. This confirms the evidence from animal experiments.

The aortic chemoreceptors are not excluded as the receptor site, and indeed the earlier reduction in heart rate than in ventilation in many instances would fit in with this explanation.

ii) medullary centres, or other sites in the brain. The delay time would be compatible with such a site of action only if a step change in oxygen concentration is more rapidly effective in the medulla than is a step change in CO_2 concentration, since for CO_2 it can be shown that central effects take definably longer than arterial chemoreceptor



Pulmonary vascular response to short-term hypoxia in man. Doyle, Wilson & Warren, 1952.



Pulmonary vascular response decreased by reoxygenation (calf). From Silove & Grover, 1968

effects. Also, if the period of ventilatory depression at arterial reoxygenation is held to be due in part to uncorrected medullary hypoxia, then the delay time is inappropriate.

iii) spinal centres: the same considerations apply: the response appears to be too rapid.

iv) the heart itself: local action on the SA node by oxygenated blood reaching it via the coronary circulation - the timing is appropriate.

v) the pulmonary circulation: reduction in pulmonary vascular resistance with haemodynamic consequences affecting pressure receptors in undetermined sites.

The timing might be appropriate: Doyle et al (1952) published a record showing a steep reduction within 20 sec of switching off 10% O_2; Silove & Grover (1967) showed a reduction in P.A.P. within 20 sec of reoxygenation in calves. No other data on timing of this response seem to be available. (Fig 5:36)

vi) other chemoreceptor site, unknown, and no further from the heart than the known ones.

vii) the peripheral vascular bed: the change would appear to be too rapid and abrupt to associate with haemodynamic adjustments following peripheral reoxygenation.

viii) the adrenal medulla - either direct reoxygenation or reduction of secretion via CNS reoxygenation would appear to require a longer time delay.

b) Effector pathways

If the tachycardia is neurally controlled and if the spinal and normal subjects respond by the same mechanism, then either;

i) the tachycardia is due to sympathetic stimulation, and the action of hypoxia is either directly or reflexly on SPINAL centres

ii) the tachycardia is due to vagal inhibition, and the action of hypoxia is directly or reflexly on medullary centres.

Animal evidence is in favour of sympathetic stimulation. This is also the pathway for response to increases in left atrial pressure, and for the inspiratory rise in heart rate. The efferent pathway for any aortic chemoreflex there may be is thought to be sympathetic, and certainly the afferents are thought to travel to the medulla. Possibly the usual response is mediated by both sympathetic stimulation and vagal inhibition, and the persistent vagal activity in the spinal subject is relatively greater than the normal vagal component.

Further experiments which might show light on these problems include:

In man: a similar air → 10% oxygen → 100% oxygen sequence with

(1) continuous monitoring of blood pressure by intra-arterial catheter in order to define the timing of any changes in relation to changes in heart rate. (This has been done on one occasion - see p 5/98)

(2) Continuous monitoring of pulmonary artery pressure.

(3) Frequent measurements of forearm blood flow, to define the timing of peripheral vascular effects.

In man, monkeys, or dogs:

Inhaled or intravenous radioisotope studies, with detectors at (a) carotid (b) medulla (c) spinal cord (d) forearm muscle. The practical difficulties here are considerable: such an experiment might however be possible by cooperation with surgical research teams already studying cerebral blood flow, and performing laminectomy.

Ideally, if (say) radioactive xenon inhalation could be used, a single breath could be combined with the first breath of oxygen at reoxygenation,

so that heart rate changes could be recorded, concurrently with appropriate circulation times.

In any such experiments, it would be desirable to monitor end-tidal oxygen concentration continuously, and where possible arterial PO_2 would also be measured.

In conclusion:

1) The carotid chemoreceptors are not responsible in man for the tachycardia of hypoxia. Aortic chemoreceptors may be.
2) The tachycardia of hypoxia is not secondary to the ventilatory response.
3) Direct or reflex action in the medulla, and its mediation by the sympathetic is not the mechanism, or not the ONLY mechanism for producing hypoxic tachycardia in man, since the response is normal in subjects with cervical spinal transection. Either the direct or reflex action of hypoxia is spinal, or the efferent pathway from the medulla is wholly or partly vagal.
4) A direct action of hypoxia on the pacemaker is not excluded.
5) An action secondary to pulmonary vasoconstriction is a further possibility which does not appear to have been suggested.

REFERENCES

Abramson, D.I., Landt, H. & Benjamin, J.E. (1943). Peripheral vascular response to acute anoxia. Arch. Intern. Med. 71: 583-593.

Alexander, R.S. (1945). The effects of blood flow and anoxia on spinal cardiovascular centers. Amer. J. Physiol. 143: 698-708.

Alveryd, A. & Brody, S. (1947). Cardiovascular and respiratory changes in man during oxygen breathing. Acta physiol. scand. 15: 140-149

Asmussen, E. & Chiodi, H. (1941). The effect of hypoxemia on ventilation and circulation in man. Am. J. Physiol. 132: 426-436.

Aviado, D.M. (1960). The Pharmacology of the pulmonary circulation. Pharm. Rev. 12: 159-232,

Barcroft, J. (1925). The Respiratory Function of the Blood, Part 1, "Lessons from High Altitudes", p.111. Cambridge: Cambridge University Press.

Bernthal, T., Greene, W. & Revzin, A.M. (1951). Role of Carotid Chemo receptors in Hypoxic Cardiac Acceleration. Proc. Soc. Exp. Biol. Med. 76: 121-124.

Bird, A.D. & Telfer, A.B.M. (1965). Effect of hyperbaric oxygen on limb circulation. Lancet 7, 355.

Biscoe, T.J. & Purves, M.J. (1965). Carotid chemoreceptor and cervical sympathetic activity during passive hind-limb exercise in the anaesthetised cat. J. Physiol., Lond. 177: 72P.

Biscoe, T.J. & Purves, M.J. (1967). Observations on the rhythmic variation in the cat carotid body chemoreceptor activity which has the same period as respiration. J. Physiol. 190: 389-412.

Biscoe, T.J. & Purves, M.J. (1967). Observations on carotid body chemoreceptor activity and cervical sympathetic discharge in the cat. J. Physiol. 190: 413-424.

Biscoe, T.J. & Purves, M.J. (1967). Factors affecting the cat carotid chemorecptor and cervical sympathetic activity with special reference to passive hind-limb movements. J. Physiol. 190: 425-441.

Black, A.M.S., McCloskey, D.I. & Torrance, R.W. (1966). The responses of peripheral chemoreceptors to sudden changes of hypercapnic and hypoxic stimuli. J. Physiol. 185: 67P.

Black, J.E. & Roddie, I.C. (1958). The mechanism of the changes in forearm vascular resistance during hypoxia. J. Physiol. 143: 226-235.

Bouverot, P., Flandrois, R. & Grandpierre, R. (1961). A propos du mechanisme d'action chemoreflexe ou central du "stimulus CO_2" de la ventilation. Compt. rend. acad. sci. Paris. 252: 790-792.

Bouverot, P., Flandrois, R., Puccinelli, R. & Dejours, P. (1963). Etude du stimulus CO_2 de la ventilation a differents niveaux d'oxygenation chez le chien eveille. J. Physiol. Paris. 55: 206-207.

Bristow, J.D., Brown, E.B., Cunningham, D.J.C., Goode, R.C., Howson, M.G. & Sleight, P. (1968). The influence of ventilation, carbon dioxide and hypoxia on the baroreceptor reflex in man. J. Physiol. 198: 102-103P.

Brooks, C.McC., Kao, F.F.,&Lloyd, B.B. (1965). Cerebrospinal fluid and the regulation of ventilation. Blackwell, Oxford.

Campbell, E.J.M. & Howell, J.B.L. (1962). Rebreathing method for measurement of mixed venous PCO_2. Brit. Med. J. ii 1962 ii 630-633.

Chalazonitis, N.A. (1969). Stimulation and depression of neurones by change in gas partial pressures and in pH. J. Physiol. 202 (1) 2P. Proceedings of Royal Postgraduate Medical School Meeting (Sympcsium on control of ventilation, Paper 3). Feb. 1969.

Christiansen, J., Douglas, C.G. & Haldane, J.S. (1914). The absorption and dissociation of carbon dioxide by human blood. J. Physiol. 48: 244-271.

Clynes, M. (1960). Respiratory sinus arrhythmia: laws derived from computer simulation. J. Appl. Physiol. 15 (5): 863-874.

Comroe, J.H. (1939). The location and function of the chemoreceptors of the aorta. Amer. J. Physiol. 127: 176-191.

Comroe, J.N. Jnr. (1964). The peripheral chemoreceptors. In Handbook of Physiology, Section 3: Respiration, Volume 1. Ed. Fenn, W.O. & Rahn, H. American Physiol. Soc. Washington, D.C.

Comroe, J.H. Jnr. & Mortimer, L. (1963). Cardiovascular and respiratory effects of sodium cyanide, nicotine, phenyldiguanide and serotonin on the aortic chemoreceptors, separated temporally from the carotid bodies. Pharmacologist 5(2) 259, 1963.

Comroe, J.H. Jnr. & Schmidt, C.F. (1938). The part played by reflexes from the carotid body in the chemical regulation of respiration in the dog. Amer. J. Physiol. 121: 75-97.

Cozine, R.A. & Ngai, S.H. (1967). Medullary surface chemoreceptors and regulation of respiration in the cat. J. appl. Physiol. 22(1): 117-121.

Cross, C.E., Rieben, P.A., Barron, C.I. & Salisbury, P.F. (1963). Effects of arterial hypoxia on the heart and circulation: an integrative study. Amer. J. Physiol. 205: 963-970.

Cunningham, D.J.C., Cormack, R.S., O'Riordan, J.L.H., Jukes, M.G.M. & Lloyd, B.B. (1957). An arrangement for studying the respiratory effects in man of various factors. Quart. J. exp. Physiol. 42: 294-303.

Cunningham, D.J.C., Elliott, D.H., Lloyd, B.B., Miller, J.P. & Young, J.M. (1965). A comparison of the effects of oscillating and steady alveolar partial pressures of oxygen and carbon dioxide on the pulmonary ventilation. J. Physiol. (1965) 179: pp.498-508.

Cunningham, D.J.C. & Lloyd, B.B. (1963). The Regulation of Human Respiration. Blackwell Scientific Publications, Oxford.

Cunningham, D.J.C., Lloyd, B.B., Miller, J.P. & Young, J.M. (1965). The time course of human ventilation after transient changes in P_ACO_2 at two levels of P_AO_2. J. Physiol. 179: 68-70P.

Daly, W.J. & Bondurant, S. (1962) Circulatory effects of oxygen breathing. Aerospace Med. 33:333

Daly, M. de B. & Scott, Mary J. (1962). An analysis of the primary cardiovascular reflex effects of stimulation of the carotid body chemoreceptors. J. Physiol. Lond. 162: 555-573.

Daly, M. de B. & Ungar, A. (1966). Comparison of the reflex responses elicited by stimulation of the separately perfused carotid and aortic body chemoreceptors in the dog. J. Physiol. 182: 379-403.

Davies, C.T.M. & Neilson, J.M.M. (1967). Sinus arrhythmia in man at rest. J. appl. Physiol. 22(5): 947-955.

De Castro, F. (1951). Sur la structure de la synapse dans les chemorecepteurs. Leur mecanisme d'excitation et role dans la circulation sanguine locale. Acta physiol. scand. 22: 14-43.

Dejours, P. (1957). Interet methodologique de l'etude d'un organisme vivant a la phase initiale de rupture d'un equilibre physiologique. Comptes rendus des seances de l'Academie des Sciences, 245: 1946-1948.

Dejours, P. (1962). Chemoreflexes in breathing. Physiol. Rev. 42 No. 3. 335-358.

Dejours, P. (1963). Control of Respiration by arterial chemoreceptors. Ann. N.Y. Acad. Sci. 109 Article 2, 682-695.

Dejours, P., Labrousse, Y., Raynaud, J. & Flandrois, R. (1958) Etude du stimulus gaz carbonique de la ventilation chez l'homme. J. Physiol. (Paris) 50: 239-243.

Dejours, P., Labrousse, Y., Raynaud, J., Girard, F. & Teillac, A. (1958). Stimulus oxygene de la ventilation au repos et au cours de l'exercice musculaire, a basse altitude (50m), chez l'homme. Revue Francaise d'etudes cliniques et biologiques. Vol. III No. 2: 105-123.

Downing, S.E. (1966). Autonomic influences on cardiac function in systemic hypoxia. Proc. int. Symp. cardiovasc. respir. Effects Hypoxia, Kingston, Ont. 1965, pp. 208-231 (Karger, Basel/New York 1966).

Downing, S.E., Mitchell, J.H. & Wallace, A.G. (1963). Cardiovascular responses to ischaemia, hypoxia and hypercapnia of the central nervous system. Amer. J. Physiol. 204: 881-887.

Downing, S.E., Remensnyder, J.P. & Mitchell, J.H. (1962). Cardiovascular responses to hypoxic stimulation of the carotid bodies. Circulat. Res. 10: 676-685.

Doyle, J.T., Wilson, J.S. & Warren, J.V. (1952). Pulmonary vascular responses to short term hypoxia in human subjects. Circulation 5: 263-270.

Dripps, R.D. & Comroe, J.H. Jnr. (1947). The effect of the inhalation of high and low oxygen concentrations on respiration, pulse rate, ballistocardiogram and arterial oxygen saturation (oximeter) of normal individuals. Amer. J. Physiol. Vol. 149 No. 2. 277-291.

Dutton, R.E., Hodson, W.A., Davies, D.G. & Chernick, V. (1967). Ventilatory adaptation to a step change in PCO_2 at the carotid bodies. J. Appl. Physiol. 23(2): 195-202.

Euler, Von U.S., Liljestrand, G. & Zotterman, Y. (1939). The excitation mechanism of the chemoreceptors of the carotid body. Skand. Arch. Physiol. 83:132-152.

Eyzaguirre, C. & Lewin, J. (1961). The effect of sympathetic stimulation on carotid nerve activity. J. Physiol. 159: 251-267.

Fitzgerald, R.S., Zajtchuk, J.T., Penman, R.W.P. & Perkins, J.F. Jnr. (1964). Ventilatory response to transient perfusion of carotid chemoreceptors. Amer. J. Physiol. 207: 6. 1964, 1305-1313.

Floyd, W.F. & Neil, E. (1952). The influence of the sympathetic innervation of the carotid bifurcation on chemoreceptor and baroreceptor activity in the cat. Arch. int. Pharmacodyn Ther. 91: 230-239.

Gelfand, R., Lambertsen, C.J. & Davies, J. (1964). Use of CO_2-induced respiratory transients in analysis of respiratory control mechanisms. The Physiologist 7: 141.

Gernandt, B.E. (1946). A study of the respiratory reflexes elicited from the aortic and carotid bodies. Acta Physiol. Scand. 11 (Suppl. 35): 1-81

Gesell, R. (1939). Respiration and its adjustments. Ann. Rev. Physiol. 1: 185-216.

Glick, G., Plauth, W.H. Jnr. & Braunwald, E. (1964). Circulatory responses to hypoxia in unanesthetized dogs with and without cardiac denervation. Amer. J. Physiol. 207: 753-758.

Gray, J.S. (1950). Pulmonary ventilation and its physiological regulation.

Greenfield, L.J. & Ebert, P.A. (1963). Cardiac denervation effect in hypoxia and hypercapnia. Effect of total denervation on the cardiovascular responses. Arch. Surg. 87; 717-721

Greer, J.R. (1958). A Sensitive Defocusing photoelectric pressure transducer. Electronic Engineering, Vol. 30 No. 365.

Greer, J.R. (1966). Integrated pneumotachograph tracings and lung volumes. World Medical Electronics, Vol. 4 No. 4.

Greer, J.R. & Jennett, Sheila (1969). An integrated pneumotachograph system including breath-by-breath measurement of ventilation rate (minute volume). J. Physiol. 200: 92-94P.

Grodins, F.S. (1964). Regulation of pulmonary ventilation. The Physiologist 7: 319-333.

Haldane, J.S. & Priestley, J.G. (1905). The regulation of the lung ventilation. J. Physiol. 32: 225-266.

Harris, P., Bishop, J.M. & Segel, N. (1961). The influence of guanethidine on hypoxic pulmonary hypertension in normal man. Clin. Sci. 21: 295-300.

Harrison, T.R. & Blalock, A. (1927). Effect of severe anoxemia of short duration on cardiac output of morphinised dop and trained unnarcotised dog. Am. J. Physiol. 80: 169.

Hatcher, J.D. & Jennings, D.B. (eds.) (1966). The cardiovascular and respiratory effects of hypoxia. S. Karger, Basel/New York 1966.

Hatcher, J.D. & Jennings, D.B. (1966). Evidence for the Role of Humoral mechanisms in the cardiovascular responses to hypoxia and anaemia. Proc. int. Symp. cardiovasc. respir. Effects Hypoxia, Kingston, Ont. 1965, 174-188.

Hernandez-Martinez, J.P. (1967). Etude experimentale du controle chemoreflexe de la frequence cardiaque Chez l'Homme au repos. Travail du Laboratoire de Physiologie de la Faculte de Medecine de Lyon (Pr. J.F. Cier).

Heymans, C., Bouckaert, J.J. & Dautrebande, L. (1931). Au sujiet du mechanisme de la bradycardie provoquee par la nicotine, la lobeline, le cyanure, le sulfure de sodium, les nitrites et la morphine, et de la bradycardie asphyxique. Arch. int. Pharmacodyn, 41: 261-289.

Heymans, J.F. & Heymans, C. (1927). Sur les modifications directes et sur la regulation reflexe de l'activite du centre respiratoire de la tete isolee du chien. Arch. int. Pharmacodyn. 33: 273-377.

Heymans, C. & Neil, E. (1958). Reflexogenic Areas of the Cardiovascular System. Churchill, London.

Holton, Pamela & Wood, J.B. (1965). The effects of bilateral removal of the carotid bodies and denervation of the carotid sinuses in two human subjects. J. Physiol. 181: 365-378.

Hornbein, T.F., Roos, A. & Griffo, Z.J. (1961). Transient effect of sudden mild hypoxia on respiration. J. Appl. Physiol. 16: 11-14.

Howell, J.B.L. & Campbell, E.J.M. (Eds.) (1966). Breathlessness. Blackwell Scientific Publications, Oxford.

Jennett, Sheila (1968). Assessment of respiratory effects of analgesic drugs. Brit. J. Anaesth. 40: 746-756.

Jennett, Sheila (1969). Changes in ventilation and heart rate during and after brief moderate hypoxia in resting man. J. Physiol. 200: 106-107P.

Joels, N. & Neil, E. (1961). The influence of anoxia and hypercapnia separately and in combination on the chemoreceptor impulse discharge. J. Physiol. 155: 45-46P.

Joels, N. & Neil, E. (1963). The excitation mechanism of the carotid body. Brit. med. Bull. 19: 21-24.

Johnson, R.H., Crampton Smith, A. & Spalding, J.M.K. (1969). Blood pressure response to standing and to Valsalva's manoeuvre: independence of the two mechanisms in neurological diseases including cervical cord lesions. Clin. Sci. 36: 77-86.

Kety, S.S. & Schmidt, C.F. (1948). Effects of altered arterial tensions of carbon dioxide and oxygen on cerebral blood flow and cerebral oxygen consumption of normal young men. J. Clin. Invest. 27: 484-499.

Korner, P.I. (1959). Circulatory adaptations in hypoxia. Physiol. Rev. 39: 687-730.

Lambertsen, C.J. (1963). Physiological Effects of oxygen. in Proc. Second Symposium on Underwater Physiology Ed. Lambertson, C.J. & Greenbaum, L.J.G. Natl. Acad. Sci. Natl. Research Council, Washington, D.C.

Lambertsen, C.J., Gelfand, R. & Kemp, R.A. (1965). Dynamic response characteristics of several CO_2-reactive components of the respiratory control system. In: Cerebrospinal fluid and the regulation of ventilation. pp. 211-240. Ed. Brooks, C. McC., Kao, F.F. & Lloyd, B.B. Blackwell, Oxford.

Lambertsen, C.J., Hall, P., Wollman, H. & Goodman, M.W. (1963). Quantitative interactions of increased PO_2 and PCO_2 upon respiration in man. Ann. N.Y. Acad. Sci. Vol. 109, Art. 2. 731-741.

Lambertsen, C.J., Kough, R.H., Cooper, D.Y., Emmel, G.L., Loeschke, H.H. & Schmidt, C.F. (1953). Comparison of relationship of respiratory minute volume to PCO_2 and pH of arterial and internal jugular blood in normal man during hyperventilation produced by low concentrations of CO_2 at 1 atmosphere and by O_2 at 3.0 atmospheres. J. Appl. Physiol. 5: 803-813.

Lambertsen, C.J. & Wendel, H. (1960). An alveolar PCO_2 control system: its use to magnify respiratory depression by meperidine. J. Appl. Physiol.15 43-48.

Landgren, S. & Neil, E. (1951). Chemoreceptor impulse activity following haemorrhage. Acta physiol. scand. 23: 158-167.

Landmesser, C.M., Cobb, S., Peck, A.S. & Converse, J.G. (1957). Respiratory responses to carbon dioxide "transients" in normal volunteers. Anesthesiology 18: 807-830.

Ledsome, J.R. & Linden, R.J. (1967). The effect of distending a pouch of the left atrium on the heart rate. J. Physiol. 193: 121-129.

Leitner, L-M., Pages, B., Puccinelli, R. & Dejours, P. (1965). Etude simultanee de la ventilation et des decharges des chemorecepteurs du glomus carotidien chez le chat. I. Au cours d'inhalations breves d'oxygene pur. Arch. int. Pharmacodyn. 154 No. 2. 421-426.

Leitner, L-M., Pages, B., Puccinelli, R. & Dejours, P. (1965). Etude simultanee de la ventilation et des decharges des chemorecepteurs du glomus carotidien chez le chat. II. Au cours d'inhalations breves d'anhydride carbonique. Arch. int. Pharmacodyn. 154: No. 2. 427-433.

Lloyd, B.B. & Cunningham, D.J.C. (1963). A quantitative approach to the regulation of human respiration. In: The Regulation of Human Respiration, 331-349. Ed. Cunningham, D.J.C. & Lloyd, B.B. Blackwell Scientific Publications, Oxford 1963.

Lloyd, B.B., Jukes, M.G.M. & Cunningham, D.J.C. (1958). The relation between alveolar oxygen pressure and the respiratory response to CO_2 in man. Quart. J. exp. Physiol. 43: 214-227.

Loeschcke, H.H., Mitchell, R.A., Katsaros, B.,Perkins, J.F. Jr. & King, A. (1963). Interaction of intracranial chemosensitivity with peripheral afferents to respiratory center. Amer. N.Y. Acad. Sci. 109: 651-660.

Lutz, B.R. & Schneider, E.C. (1919). Circulatory responses to low oxygen. Amer. J. Physiol. 50: 228.

Mathison, G.C. (1910). The Action of Asphyxia upon the Spinal animal. Jour. Physiol. xli: 416-449.

Meyer, J.S., Gotch, F. & Takagi, Y. (1967). Inhalation of Oxygen and Carbon Dioxide Gas. Effect on composition of cerebral venous blood. Arch. int. Med. 119: 4-15.

Mitchell, R.A., Loeschcke, H.H., Massion, W.H. & Severinghaus, J.W. (1963). Respiratory responses mediated through superficial chemosensitive areas on the medulla. J. appl. Physiol. 18: 523-533.

Mitchell, R.A. (1965). The regulation of respiration in metabolic acidosis and alkalosis. in Cerebrospinal fluid and the regulation of ventilation: Blackwell, Oxford. Brooks, C.McC., Kao, F.F. & Lloyd, B.B. (Eds.).

Mitchell, R.A., Loeschcke, H.H., Severinghaus, J.W., Richardson, B.W. & Massion, W.H. (1963). Regions of respiratory chemo-sensitivity on the surface of the medulla. Ann. N.Y. Acad. Sci. 109: 661-681.

Motley, H.L., Cournand, A., Werke, L., Himmelstein, A. & Dresdale, D. (1947). The influence of short periods of induced acute anoxia upon pulmonary artery pressures in man. Am. J. Physiol. 150: 315-320.

Nahas, G.G. (Ed.). (1963). Regulation of Respiration. Ann. N.Y. Acad. Sci. Vol. 109. Art. 2. 411-948.

Neil, E. (1956). Influence of carotid chemoreceptor reflexes on heart rate in systemic anoxia. Arch. int. Pharmacodyn. 105: 477-488.

Neil, E. (1966). In: Cardiovascular and Respiratory Effects of Hypoxia. Ed. Hatcher, J.D. & Jennings,D.B. p. 170.

Neil, E. & Joels, N. (1963). The carotid glomus sensory mechanism. In: The regulation of human respiration. 163-171. Ed. Cunningham, D.J.C. & Lloyd, B.B. Blackwell, Oxford.

Neilsen, M. (1936). Untersuchungen über die Atem regulation beim Menschen. Skand. Arch. Physiol. 74, Suppl. 10: 83-208.

Neilsen, M. & Smith, H. (1951). Studies on the regulation of respiration in acute hypoxia. Acta physiol. scand. 24: 293-313.

Noble, M.I.M., Trenchard, D. & Guz, A. (1967). Effect of changes in P_aCO_2 and P_aO_2 on cardiac performance in conscious dogs. J. appl. Physiol. 22(1): 147-152.

Opitz, E. (1941). Ueber akute Hypoxie. Ergeb Physiol. Exptl. Pharmacol. 44: 315-424.

Pickering, G.W., Sleight, P. & Smyth, H.S. (1968). The reflex regulation of arterial pressure during sleep in man. J. Physiol. 194: 46-48P.

Pollock, J.G., Bell, G. & Ledingham, I. McA. (1968). The response of the peripheral circulation in man to varying tensions of inspired oxygen, as measured by strain gauge plethysmography and the clearance of Xenon^{133} from muscle. In: Blood Flow through Organs and Tissues. p. 408-415. Ed. Bain, W.H. & Harper, A.M. E. & S. Livingstone, LA, Edinburgh and London.

Purves, M.J. (1965). Oscillations of oxygen tension in arterial blood. J. Physiol. 176: 7-8P.

Purves, M.J. (1966). Fluctuations of arterial oxygen tension which have the same period as respiration. Resp. Physiol. 1: 281-296.

Read, D.J.C. & Leigh, J. (1967). Blood-brain tissue PCO_2 relationships and ventilation during rebreathing. J. Appl. Physiol. 23: 53-70.

Reed, D.J. & Kellog, R.H. (1960). Effect of sleep on hypoxic stimulation of breathing at sea level and altitude. J. Appl. Physiol. 15(b):1130-1134.

Reed, D.J. & Kellog, R.H. (1960). Effect of sleep on CO_2 stimulation in acute and chronic hypoxia. J. Appl. Physiol. 15(b): 1135-1138.

Resnik, W.H. (1925). Observations on the effect of anoxemia.on the heart. III. J. clin. Invest. 2: 125-141.

Richardson, D.W., Kontos, H.A., Raper, A.J. & Patterson, J.L. (1967). Modification by Beta-Adrenergic blockade of the circulatory responses to acute hypoxia in man. J. clin. Invest. 46(1): 77-85.

Richardson, D.W., Kontos, H.A., Shapiro, W. & Patterson, J.L. (1966). Role of hypocapnia in the circulatory response to acute hypoxia in man. J. appl. Physiol. 21: 22-36.

Riedstra, J.W. (1963). Influence of central and peripheral PCO_2 (pH) on the ventilatory response to hypoxic chemoreceptor stimulation. Acta physiol. pharmacol. neerl. 12: 407-452.

Sands, Jane & De Graff, A.C. (1925). The effects of Progressive Anoxemia on the Heart and Circulation. Amer. J. Physiol. 74(2): 416-435.

Schaefer, K.E. (1958). Respiratory pattern and respiratory response to carbon dioxide. J. Appl. Physiol. 13: 1-14.

Scott, M.J. (1959). The Reflex Control of the Circulation by Chemoreceptors. Ph.D. Thesis London.

Severinghaus, J.W., Chiodi, H., Eger, E.I., Braudelater, B. & Hornbein, T.F. (1966). Cerebral Blood Flow in Man at High Altitude. Circulation Research, Vol.XIX, 274-282.

Silove, E.D. & Grover, R.F. (1968). Effects of Alpha Adrenergic Blockade and Tissue catecholamine depletion on Pulmonary vascular Response to Hypoxia. J. clin Invest. 47(2): 274-285.

Shock, N.W. & Soley, M.H. (1940). Effect of breathing pure oxygen on respiratory volume in humans. Proc. Soc. Exp. Biol. Med. 44(2): 418-420.

Torrance, R.W. (1968). Prolegomena. p. 1-40. In: Arterial Chemoreceptors. Ed. Torrance, R.W. Blackwell, Oxford.

Vogel, J.A. & Harris, C.W. (1967). Cardiopulmonary responses of resting man during early exposure to high altitude. J. appl. Physiol. 22(6): 1124-1128.

Wade, J.G., Larson, C.P., Hickey, R.H. & Ehrenfild, W.K. (1969). Effect of carotid endarterectomy on carotid chemoreceptor and baroreceptor function. Surg. Forum 144-145.

Watt, J.G., Dumke, P.R. & Comroe, J.H. Jnr. (1943). Effects of inhalation of 100% and 14% oxygen upon respiration of unaesthetised dogs before and after chemoreceptor denervation. Amer. J. Physiol. 138: 610-617.

Winterstein, H. (1961). The actions of substances introduced into the cerebrospinal fluid and the problem of intracranial chemoreceptors. Pharmac. Rev. 13: 71-107.

Yamamoto, W.S. & Edwards, M.W. (1960). Homeostasis of CO_2 during IV infusion of CO_2. J. Appl. Physiol. 15: 807-818.

Sheila Jennett SUMMARY Ph.D.

HYPOXIA AND HYPEROXIA IN MAN: IMMEDIATE EFFECTS ON VENTILATION AND HEART RATE

Three problems related to ventilatory and cardiovascular control in resting man have been studied; emphasis was placed on the timing of response to changes in concentration of inspired gases, and thus on the assessment of the part played by arterial chemo-reflexes. Observations were made on the immediate effects, by continuous recording of ventilatory parameters, and in many instances of heart rate also.

1. The response of ventilation to increase in inspired CO_2 concentration during hyperoxia.

   This was studied in 9 normal subjects.

   Ventilation was recorded during a series of 2-breath tests in which 7% CO_2 was substituted briefly for the background of air, or of oxygen respectively.

   In a few subjects only, there was evidence of an arterial chemoreflex response in hyperoxia.

2. The changes in ventilation and in heart rate during the first few minutes of breathing 100% oxygen.

   Ventilatory responses were studied in 11 normal subjects, and heart rate also in 3.

   Initial transient depression of ventilation was confirmed in many instances: more particularly timing of the secondary stimulation was studied; and it was found to develop most often in the second minute. Bradycardia, when it occurred developed more gradually from the first half minute.

3. The tachycardia induced by hypoxia, and its relationship to ventilatory changes. This was the major problem studied, and the largest section of the thesis is devoted to it.

The mechanism of the tachycardia of hypoxia in man is not known: earlier evidence has only partly supported the most likely explanation from animal studies, namely that central hypoxic stimulation leads to increased sympathetic activity and thence to neural pacemaker stimulation.

Subjects were exposed to moderate brief hypoxia (10% oxygen for 3 - 10 min); this was abruptly relieved by 100% oxygen. The changes at reoxygenation were observed particularly closely. The following were studied:

i) 6 normal men, repeatedly, in three different series of experiments.

ii) 8 patients with complete transection of the cervical spinal cord.

iii) A patient following bilateral removal of carotid body tumours.

In all these subjects, heart rate increased during hypoxia, and decreased to normal abruptly after a delay of 10-18 sec from the beginning of the first breath of oxygen. The changes were not linked to changes in ventilation; this was observed on many individual occasions, and was conclusively established by a series of experiments in which normal subjects were trained to maintain their breathing constant during the period of the change from 10% to 100% oxygen: the heart rate nevertheless decreased and after a similarly short delay.

The normality of the heart rate changes in the

patients with cervical cord transection indicates that medullary stimulation and increased sympathetic efferent activity is not the mechanism, (or not the only mechanism), responsible for hypoxic tachycardia in man. The timing of the response is consistent with an arterial chemoreflex: the normal response in the patient with absent carotid body function leaves open the possibility of the aortic bodies as the receptors. This and other possibilities, such as a link with pulmonary vascular changes, are discussed, and further human and animal studies suggested, to elucidate the cause of the tachycardia.