NON-INVASIVE METHODS OF MEASURING CARDIAC OUTPUT DURING EXERCISE

A thesis submitted by

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for the degree of Doctor of Philosophy
at the University of London.

August, 1974 .

ABSTRACT

This thesis investigates the reproducibility and reliability of several non-invasive CO_2 and N_2O rebreathing methods of measuring cardiac output in normal subjects during exercise.

In the CO_2 rebreathing method, the dependance of cardiac output on the value of mixed venous PCO_2 ($P\bar{v}CO_2$) was initially assessed in chapter 3. A comparison of the exponential and plateau (or it's linear extrapolation) rebreathing methods indicated that the latter method was more reliable. Over a wide range of CO_2 rebreathing concentrations the linearly extrapolated values of $P\bar{v}CO_2$ (and their variability) did not differ markedly from plateau $P\bar{v}CO_2$ estimates. In contrast, exponentially extrapolated values of $P\bar{v}CO_2$ were affected by the CO_2 concentration of the initial rebreathing mixture, and their variability was reduced only when the CO_2 content of the rebreathing mixture was raised to levels adequate for a plateau $P\bar{v}CO_2$ determination.

The plateau CO_2 rebreathing method was subsequently used to determine the day-to-day variation in $P\bar{v}CO_2$ and cardiac output (chapter 4). The mean intra-subject variation of $P\bar{v}CO_2$ during exercise (C.V of 2-3%) was 1-2% greater than the variation measured on a single occasion by previous workers. The values of cardiac output and their variability (intra-subject C.V of 22% at rest, and 5-6% during exercise) were also similar to those found by previous workers using invasive methods.

In chapter 5 an investigation of the Ayotte (1970) and the modified Cander and Forster (1959) N_2O rebreathing methods indicated that they under-estimated cardiac output, and were poor in reproducibility.

Their major fault was an assumption of a fixed lung-bag rebreathing volume, which under-estimated the amount of N_2O absorbed by pulmonary blood. In the Ayotte method, further errors in the measurement of the mean alveolar N_2O concentration probably caused a further under-estimation of cardiac output.

The N_2O rebreathing method was modified to eliminate possible sources of error (chapter 2c). Using the modified method, the absolute values and variability of cardiac output were comparable to those based on direct invasive methods (chapter 6).

A comparison of almost simultaneous estimates of cardiac output using the CO_2 and N_2O rebreathing methods showed them to be comparable in reproducibility during exercise, while absolute values of cardiac output were generally within a $\pm$ 20% line of identity (chapter 7).

The dance along the artery

The circulation of the lymph

Are figured in the drift of stars.....

.....And the end of all our exploring

Will be to arrive where we started

And know the place for the first time.

T. S. Eliot (Four Quartets)

DEDICATION

This thesis is dedicated to the many people who have contributed towards its making. I hope that it will be of service to those workers who introduced the notion that "rest" is a four-letter word.

A C K N O W L E D G E M E N T S

The work for this thesis was carried out in the Department of Pediatrics at the Cardiothoracic Institute, and in the Respiratory Physiology Unit at Hammersmith Hospital. It was supported by grants from the Brompton Hospital and the Medical Research Council. The completion of the work was possible due to the assistance of many people who combined kind thoughts with their deeds.

Sincerest thanks are extended to my supervisor Dr. Simon Godfrey, who has always tempered the demands of clear thinking in research with a patience of character.

At the Cardiothoracic Institute Doctors Anderson, Silverman, Taylor and Light were always willing to lend their experience, ideas and enthusiasm to the work. In their presence the pediatric laboratory was more aptly called an industry.

A great debt is owed to the Respiratory Physiology Unit at Hammersmith Hospital for extending their facilities and expertise to later parts of this work. In thanking Professor Fletcher and Doctors Pride, Edwards and Hughes, I also thank the members of their departments who were always available when problems of work arose.

Mr. E. E. Davies was an invaluable presence throughout this period. His knowledge and sweat in making the machinery operate is evidenced in the fact that actual results are presented in this work; the individual numbers, however, represent the subjects who volunteered to do an innumerable number of exercise tests. The fluid balance of all these

colleagues was never adequately re-adjusted in the few hours remaining after each working day.

The appearance of the thesis is due to the devotion of several workers. Miss K. Patel was the artistic flair behind the figures. Mrs. M. Strugnell, Miss R. Mannoring and Mrs. S. Abrams provided the script and tables; their work is distilled in a choice variety of three typescripts which appear in various parts of the text.

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ABBREVIATIONS

Physical parameters:

M.W	Molecular weight
O_2	Oxygen
CO_2	Carbon dioxide
N_2	Nitrogen
A	Argon
N_2O	Nitrous oxide
NO	Nitric oxide
CO	Carbon monoxide
Ne	Neon
S.T.P.D.	Standard temperature and pressure dry
A.T.P.S.	Ambient temperature and pressure saturated

Cardio-vascular parameters:

$PaCO_2$	Pressure of carbon dioxide in arterial blood
P_ECO_2	Pressure of carbon dioxide in mixed expired air
$P_{ET}CO_2$	Pressure of carbon dioxide in end tidal breath
$\bar{P}vCO_2$	Pressure of carbon dioxide in mixed venous blood
PaO_2	Pressure of oxygen in arterial blood
$\bar{P}vO_2$	Pressure of oxygen in mixed venous blood
$CaCO_2$	Content of carbon dioxide in arterial blood
$C\bar{v}CO_2$	Content of carbon dioxide in mixed venous blood
$\dot{Q}$	Cardiac output
$\dot{Q}_C$	Pulmonary blood flow
$\dot{Q}N_2O$	Cardiac output measured by the nitrous oxide rebreathing method
$\dot{Q}CC_2$	Cardiac output measured by the carbon dioxide rebreathing method
$\dot{Q}O_2$	Cardiac output measured by the direct (O_2) Fick method
$\dot{Q}$dye	Cardiac output measured by the dye dilution method
E.C.G.	Electro-cardiograph
H.R.	Heart rate
S.V	Stroke volume
pH	pH

Ventilatory parameters:

$\dot{V}_E$	Expired ventilation
$\dot{V}_D$	Physiological dead space
V_T	Tidal volume
f	Breathing frequency
$\dot{V}O_2$	Oxygen consumption
$\dot{V}CO_2$	Carbon dioxide output
$\dot{V}N_2O$	Nitrous oxide uptake
$F\bar{A}N_2O$	Fractional concentration of mean alveolar nitrous oxide
FiN_2O	Fractional concentration of inspired nitrous oxide
FfN_2O	Fractional concentration of nitrous oxide expired in the final breath
FiNe	Fractional concentration of inspired neon
FfNe	Fractional concentration of neon expired in the final breath
FAN_2O	Fractional concentration of alveolar nitrous oxide
λTISS	Solubility constant of nitrous oxide in lung tissue
λB	Solubility constant of nitrous oxide in human blood

Statistical parameters:

$\bar{x}$ or $\bar{m}$	The mean or average
S.D	Standard deviation
S.Dd	Standard deviation of the difference
C.V	Coefficient of variation
C.V.P.E.	Coefficient of variation derived from paired estimates

CHAPTER 1

INTRODUCTION

HISTORY

In 1870 an academician by the name of Adolph Fick proposed a method for measuring cardiac output from the exchange of a gas such as oxygen or carbon dioxide across the lung (Fick, 1870). Cardiac output was calculated from the rate of gas exchange, divided by the veno-arterial (or arterio-venous) content difference of the gas in blood.

In terms of carbon dioxide:

$$\dot{Q}CO_2 \text{ (l/min)} = \frac{\dot{V}CO_2}{C\bar{v}CO_2 - CaCO_2}$$

where $\dot{Q}CO_2$ = cardiac output in litres/minute

$\dot{V}CO_2$ = carbon dioxide output

$C\bar{v}CO_2$ = content of carbon dioxide in mixed venous blood

$CaCO_2$ = content of carbon dioxide in arterial blood

The Fick principle became the foundation of most methods of measuring cardiac output. Unfortunately, however, it necessitated the use of invasive catheterisation techniques which did not come into routine use until the early 1940's (Cournand, 1945; Hamilton, 1945), when more accurate methods of measuring blood gases became available. Thus, although the Fick principle for measuring cardiac output was validated in studies on dogs (Grehant and Quinquad, 1886) it could not readily be applied to studies of human subjects.

MEASUREMENT OF CARDIAC OUTPUT USING CARBON DIOXIDE

At the turn of the century physiologists were faced with the problem of measuring cardiac output without the availability of catheterisation techniques. The most difficult measurement was that of the mixed venous PCO_2 (and PO_2). Several workers therefore attempted to measure it through the analysis of respired gas. The lung was used as an aerotonometer in order to establish an equilibrium between the alveolar and mixed venous blood concentration of a physiological gas.

The use of the lung as an aerotonometer was first attempted in laboratory animals by Wolffberg (1871), who occluded a bronchus to demonstrate an equilibrium between the carbon dioxide tensions of trapped air and mixed venous blood. The method was subsequently used by Lowy and Schrotter (1903) who obstructed a second or third order human bronchus using a silver catheter, to demonstrate a similar equilibrium between the gas tensions of trapped air and mixed venous blood.

Bronchoscopy was obviously a tedious procedure which was inapplicable as a general technique. Further developments were therefore aimed at using the whole lung as an aerotonometer, with the mouth as the sampling site for all respired gases. Two methods were evolved, both of which were basically breathholding techniques. The first method was a rebreathing procedure which trapped the lung volume in a slightly larger lung-bag rebreathing volume. In the second method, a prolonged single expiration allowed the gas content of the trapped lung volume to be measured over an extended period of time.

Plesch (1909) used a rebreathing procedure to measure the mixed venous gas tensions of PCO_2 and PO_2 indirectly from samples taken at the mouth. Using the single-expiration procedure, Christensen et al. (1914) were able to measure the in-vivo dissociation curve of carbon dioxide in blood. The method was used by Henderson and Prince (1917) to measure the mixed venous PCO_2 during exercise, with reasonable success. Douglas and Haldane (1922) also measured mixed venous PCO_2 during exercise, and used the value to estimate cardiac output by the Fick equation (The Indirect Fick CO_2 method). Even by today's standards their estimates were fairly accurate.

Research into the various non-invasive methods of measuring mixed venous PCO_2 and cardiac output was diverted by the introduction and popularity of catheterisation methods. However the non-invasive concept was revived with the introduction of fast response gas analysers which were able to measure tidal and alveolar CO_2 gas tensions during normal breathing.

In 1956 Collier revived interest in the carbon dioxide rebreathing method. He found that by rebreathing an appropriate mixture of carbon dioxide in oxygen, he could achieve an equilibrium between the alveolar and mixed venous blood gas tension of CO_2 within a ten second period. This appeared as a plateau concentration of CO_2 in the mixture which was rebreathed by the subject. The plateau period lasted several seconds before recirculation of mixed venous blood ended the period of equilibrium.

Ashton and McHardy (1963) used the Collier rebreathing method to measure mixed venous PCO_2 ($P\bar{v}CO_2$) during exercise. They found that the plateau estimates of $P\bar{v}CO_2$ were reproducible, however the plateau levels were slightly affected by the initial rebreathing gas concentration. Considerable work by Jones et al. (1967), Denison (1968) and Edwards (1969) elucidated the use of the rebreathing procedure and redefined the plateau mixed venous PCO_2 as occurring between six and ten seconds during the rebreathing period. Denison (1968) and Edwards (1969) proposed the use of a linear extrapolation to estimate $P\bar{v}CO_2$ in rebreathing procedures where an equilibration plateau did not occur. Their work was subsequently confirmed by Kelman (1973) who used a computer model analysis of the rebreathing procedure.

Jones et al. (1967) found that during a rebreathing plateau period there was a difference between the $P\bar{v}CO_2$ measured at the mouth, and the PCO_2 of blood reaching the brachial artery after it had passed through the lung during the plateau period. Since the blood was measured downstream from the lung, the observation was called the "downstream effect". The observation was extended by Denison et al. (1969) who noted a similar difference between the rebreathing plateau $P\bar{v}CO_2$ and the PCO_2 of pulmonary arterial blood entering the lung (the "upstream effect").

Several explanations of the effect were put forward (Gurtner et al. 1969; Laszlo et al. 1971). One of the more

plausible explanations was proposed by Gurtner et al. (1972; 1973). They suggested the existance of a negatively charged endothelial membrane which produced an intra-capillary gradient of carbon dioxide by affecting the relative chemical reactions (and diffusion rates) of the hydrogen and bicarbonate ions in blood. The CO_2 gradient was dependant on the rates of these reactions and would be altered by the pulmonary blood flow through the capillary. However, any explanations of the "downstream" effect are still controversial.

A related controversy is whether the alveolar $P\bar{v}CO_2$ measured during the rebreathing period, or the same value corrected for the downstream effect should be used to estimate cardiac output.

Several workers (Denison et al. 1969; 1971; Godfrey et al. 1971) found that the alveolar values of $P\bar{v}CO_2$ gave the more reasonable results of cardiac output in adults and children. The results compared favourably with simultaneous direct (O_2) Fick estimates of cardiac output (Denison et al. 1969) and to values in the literature.

The calculations of cardiac output were greatly facilitated by the graphical solutions of the Fick and Bohr equations (McHardy et al. 1967). Computers further simplified the analysis of the carbon dioxide and oxygen dissociation curves (Kelman, 1966; 1967). In 1970, Godfrey introduced a computer program which was capable of calculating the results of exercise studies using the indirect (CO_2) Fick method.

Concomitant with the use of the plateau rebreathing procedure, Defares (1958) developed a method of measuring mixed venous PCO_2 from the exponential change in the alveolar PCO_2 during a rebreathing period. Various graphical solutions for the estimation of $P\bar{v}CO_2$ by this method were introduced by Defares (1958; 1972), Jernerus et al. (1963) and Klausen (1965). However the estimates of cardiac output based on these values of $P\bar{v}CO_2$ (Klausen, 1965; Ferguson et al. 1968; Clausen et al. 1970; Bar-Or et al. 1971) were shown by Godfrey and Wolf (1972) to have a wide degree of scatter.

Godfrey and Wolf (1972) compared the reproducibility of the exponential rebreathing method with that of the plateau rebreathing method and found that the latter gave more consistent estimates of both $P\bar{v}CO_2$ and cardiac output.

THE MEASUREMENT OF CARDIAC OUTPUT USING NITROUS OXIDE (OR OTHER FOREIGN GASES)

A modification of the original Fick principle was introduced by Bornstein (1910), to allow the measurement of cardiac output from the inhalation of a gas which was physically soluble in blood. The solubility of such a gas in blood is defined by Henry's law, which states:

"The solubility of a gas in a liquid is directly proportional to the pressure of the gas above the liquid at equilibrium"

When a physically soluble gas (for example nitrous oxide) is inhaled into the lung, some of the gas is absorbed by pulmonary blood; the amount absorbed is related to the partial

pressure of the gas in the lung, and to the solubility of the gas in blood. If the gas is kept in the lung over a period of time, there is a continuous though declining rate of removal of gas, as fresh volumes of blood enter the lung.

The amount of gas removed per minute (V_{GAS}) depends on the volume of blood passing through the lungs per minute ($\dot{Q}C$). It would also depend on the alveolar concentration of the gas ($F_A N_2O$), and its solubility in blood (λB), so that:

$$V_{GAS} = \dot{Q}C \times F_A N_2O \times \lambda B$$

Rearranging this, the pulmonary blood flow (cardiac output) can be calculated from the amount of gas removed from the lung:

$$\dot{Q}C = \frac{V_{GAS}}{F_A N_2O \times \lambda B}$$

The initial work of Bornstein was expanded by various workers who investigated the use of several foreign gases, including acetylene and nitrous oxide, for the measurement of cardiac output.

Krogh and Lindhard (1912) used a nitrous oxide breathholding method to measure cardiac output. The method required the collection and analysis of two alveolar samples separated by an interval of fifteen seconds. Lindhard (1923, 1925) subsequenly used the method, with nitrous oxide and ethylene as the "foreign gas" mixtures.

A rebreathing form of the Lindhard experiments was introduced by Marshall and Grollman (1928) who used ethylene and nitrous oxide to measure cardiac output. Their method required rebreathing periods of up to 30 seconds and gave fairly reproducible estimates of cardiac output. Grollman (1929) investigated the use of several gases in search of an "ideal" gas for the measurement of cardiac output; he suggested the preferential use of acetylene, with nitrous oxide as a close second, even though its analysis was tedious.

The values of cardiac output measured at rest by Grollman were later found to be low when compared with direct (O_2) Fick measurements made during catheterisation. Werko et al. (1949) and Chapman et al. (1950) compared the acetylene rebreathing method with the direct (O_2) Fick method; the latter workers found a high degree of correlation between the two methods of estimating both the arterio-venous oxygen content difference in blood, and cardiac output. However, the cardiac output based on the acetylene method was consistently 22% lower than the direct (O_2) Fick estimate. Chapman et al. (1950) also found measurable quantities of recirculated acetylene in the lung ten seconds after the start of a rebreathing period.

Christensen (1931a, 1931b, 1933) used the acetylene rebreathing method to measure cardiac output during exercise. He argued that the effect of recirculating acetylene was not significant during exercise, since a large amount of it would be

lost in the expanded vascular bed of the muscles. This argument received support from Assali and Ross (1959) who found that the partition coefficient of nitrous oxide between muscle and blood was 1.2. Other workers (Asmussen and Neilsen, 1952; Saltin, 1964a; 1964b) also used the acetylene rebreathing method during exercise and found impressive results which were comparable to dye dilution estimates of cardiac output. Saltin (1964a) recognised the existance of a consistent small error in his estimates of cardiac output, since he was unable to account for a quantity of acetylene which dissolved in the lung parenchymal tissue during the rebreathing period (Cander and Forster, 1959; Jernerus et al. 1963).

In 1959 Cander and Forster published the results of a breathholding technique for the measurement of cardiac output using several foreign gases, including acetylene and nitrous oxide. Their method measured both the recirculation time of venous blood, and the amount of "foreign" gas absorbed by the lung tissues; up to this time the volume of lung tissue which absorbed foreign gas had not been determined. The authors found that ignoring tissue uptake of gas could under-estimate cardiac output by 10%, an observation later confirmed by Jernerus et al. (1963). The estimates of lung tissue volume measured by Cander and Forster were also confirmed by Sackner et al. (1964) using a body plethysmograph technique. In 1960, Johnson et al. used the Cander and Forster breathholding method to measure cardiac output during exercise and included the correction for lung tissue absorption of gas.

Bekclake et al. (1962) introduced a steady state nitrous oxide rebreathing method for measuring cardiac output. During exercise the method compared favourably with dye dilution methods, and was moderately reproducible. However, in this method it was necessary to continue breathing a nitrous oxide mixture for up to half a minute. Despite this there was no immediate method of noting the recirculation of venous blood, the effect of which could only be estimated from the subsequent calculation of cardiac output.

Rigatto (1967) re-introduced the nitrous oxide rebreathing technique for measuring cardiac output and used mass spectrometry for all gas analyses. His method did not correct for nitrous oxide dissolved in lung tissue, which therefore produced a consistent small error in the measurement of cardiac output. The study did however indicate that heart rate and oxygen consumption during exercise were not affected by the rebreathing manouvre. The observation was of considerable significance since estimates of cardiac output at rest were generally corrected for changes in heart rate induced by the rebreathing procedure; another correction was applied for the difference between oxygen uptake measured during the rebreathing period, and the steady state oxygen consumption measured prior to it. Rigatto's study suggested that these corrections were unnecessary during exercise.

Recently, several workers (Ayotte et al. 1970; 1973; Simmons and Shephard, 1971; Pugh, 1972) have introduced novel practical techniques which have reflected both a sophistication and simplification of the foreign gas rebreathing methods for the measurement of cardiac output.

AIMS OF THE PRESENT STUDY

When the work for this thesis began there were several non-invasive rebreathing techniques in use for the measurement of cardiac output. However, in using these rebreathing methods, workers often differed in their theoretical and practical approach to the measurement of cardiac output. Despite this, no studies had adequately compared the various forms of the carbon dioxide and nitrous oxide rebreathing techniques. In addition, very few studies had investigated the reproducibility of the methods systematically on individual subjects.

The present work was carried out to study the use of some of these rebreathing methods for estimating cardiac output at rest and during exercise.

The plateau CO_2 rebreathing method (Collier, 1956) and the exponential CO_2 rebreathing method (Defares, 1956) are both used to estimate mixed venous PCO_2, which is then used to calculate cardiac output by the indirect (CO_2) Fick equation. As outlined earlier, the two methods differ markedly in their approach to the measurement of $P\bar{v}CO_2$. The reproducibility of $P\bar{v}CO_2$ measured by the two methods was therefore assessed in Chapter 3. The less variable rebreathing method was then used repeatedly on individual subjects in order to examine the reproducibility and accuracy of the calculated cardiac output (Chapter 4).

In the foreign gas rebreathing technique there is a similar proliferation of methods for the measurement of cardiac output. Most of these methods use nitrous oxide or acetylene gas, and the thecretical use of either gas is identical.

With a single exception (Becklake et al., 1967) all nitrous oxide methods involve a rebreathing (or breathholding) procedure to measure cardiac output. However, once again there are important practical differences between methods; several N_2O rebreathing methods were therefore investigated in this thesis.

The reproducibility and accuracy of the N_2O rebreathing methods of Ayotte (1970), and of Cander and Forster (1959; modified from a breathholding technique for the purposes of this study) were initially examined in Chapter 5. Possible sources of error in the two methods were eliminated in an attempt to produce a more accurate rebreathing method.

Using the modified nitrous oxide rebreathing method, the reproducibility of cardiac output measurements was re-examined in Chapter 6.

In a final study, the most reproducible carbon dioxide and nitrous oxide rebreathing procedures were used alternately to compare the measurements of cardiac output in individual subjects during exercise. (Chapter 7).

CHAPTER 2

APPARATUS AND TECHNIQUES

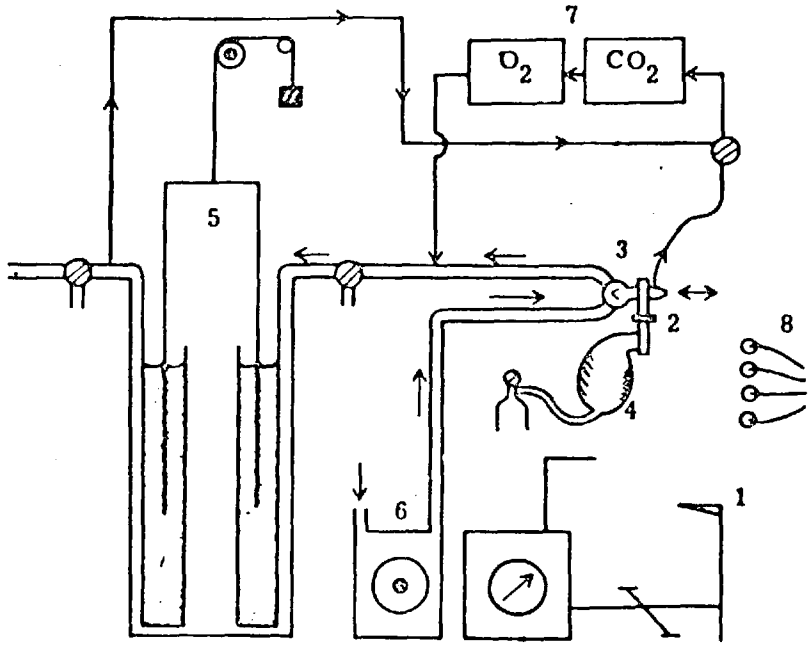
(a) THE CARBON DIOXIDE REBREATHING METHOD

All studies were carried out on normal subjects who were seated on a bicycle ergometer. A diagram of the apparatus used is shown in Figure 2.1. (Taken from Godfrey, 1971).

An electrocardiograph was used to monitor heart rate both at rest and during exercise. The subject breathed normally from a valve box attached to a rotary plate tap and mouthpiece. Inspired air was passed through a dry gas meter and expired air was passed into a large Tissot spirometer which was fitted with a mixing fan. The mixed-expired gas was either vented or collected. The concentrations of CO_2 and O_2 in the mixed expired gas were continually monitored by an infra-red CO_2 analyser and a paramagnetic oxygen analyser. The CO_2 analyser was also used to measure the end-tidal concentration of carbon dioxide at the mouth when desired.

A 5-litre open-ended anaesthetic bag was attached to one of the ports on the rotary plate, and was used for rebreathing purposes. The bag was filled with mixtures of carbon dioxide in oxygen, using an array of several gas cylinders. The subject rebreathed from the bag for periods up to 15 seconds in order to determine the mixed venous PCO_2.

All measurements of ventilation, gas concentrations, and ECG were recorded on an electronic chart recorder. Further descriptions of the apparatus and exercise test are given below.



1. Cycle 2. Tap and mouth piece 3. Valve box
4. Rebreathing bag and gas supply 5. Tissot
6. Gas meter 7. Gas analysers 8. E.C.G.

Figure 2.1 Diagram of the circuit used for the performance of exercise tests (taken from Godfrey, 1971).

THE APPARATUS

LODE BICYCLE ERGOMETER: The bicycle ergometer was an electrically braked device which presented a fixed work load to the subject over a range of pedalling frequencies.

ROTARY PLATE MOUTHPIECE AND VALVE-BOX: The rotary plate mouthpiece was made of two circular perspex plates opposed to each other, and greased to prevent leaks. One of the plates was fixed to a rigid support, and contained a single port for the mouthpiece; the mouthpiece was itself provided with a sample port for end-tidal gas analysis. The other plate could be freely rotated about the fixed plate, and was fitted with a handle attachment. Ports on this plate carried the valve-box and the rebreathing bag, either of which could be connected to the subjects mouthpiece by rotation of the plate.

The valve-box was detachable and was based on a Lloyd flap-valve design ("Hammersmith" valve). The total dead space volume of the valve box and mouthpiece was 53 ml.

GAS METER: Inspired gas volume was measured with a dry gas meter (Parkinson-Cowan CD4). The spindle of the meter (10 l. sweep) was fitted with a potentiometer. Rotation of the spindle produced a potentiometer signal which was recorded on an ink-jet recorder. The deflection of the ink-jet was linearly related to the gas meter spindle deflection. The gas-meter was calibrated using a Tissot spirometer.

TISSOT SPIROMETER: Expired gas was flushed through or collected in a 100 l. capacity Tissot spirometer. The spirometer contained a fan

to mix the collected expired gas. A rotary potentiometer was also attached to the spindle of the spirometer, so that volume displacement was recorded potentiometrically on the pen recorder (4.785 litres = 6.0 cm. deflection). The spirometer was calibrated using a water displacement method; the spirometer was then used to calibrate the dry gas meter by a gas displacement method.

RESISTANCE OF THE CIRCUIT: The internal diameter of the tubing, valve and taps was 3.8 cms. The inspiratory circuit consisted of all the pathways to the mouthpiece, including the dry gas meter; the expiratory circuit included the Tissot spirometer.

The resistance of the inspiratory and expiratory circuits was estimated with a rotameter and a variable-speed blower which generated a steady flow rate. A water manometer was used to measure the pressure generated by flow across the mouthpiece. The resistances of the circuit were found to be:

	FLOW RATE (l/min)	RESISTANCE ($cmH_2O/l/sec$)
INSPIRATION	200	0.39
	400	1.06
EXPIRATION	200	0.82
	400	1.60

INFRA-RED CO_2 ANALYSER (URAS-4): During normal breathing the mixed expired or end-tidal carbon dioxide concentrations were measured on an infra-red CO_2 analyser. The analyser was a fast response instrument with a lag-time of less than 0.5 seconds, as used in the circuit. The instrument was set to sample at a flow rate of 0.5 l/min; the sampled gas then passed into the oxygen analyser, and was finally returned

to the Tissot spirometer. During the rebreathing period the analysers sampled high concentrations of carbon dioxide and oxygen which were vented to room air to avoid subsequent contamination of mixed expired gas.

PARAMAGNETIC OXYGEN ANALYSER (SERVOMEX OA-150): The oxygen concentration of mixed expired gas was measured on a paramagnetic oxygen analyser. The analyser was a slow response instrument with a 15 second response time for full scale deflection. The instrument was used solely for the measurement of mixed expired oxygen concentration.

USE OF THE GAS ANALYSERS: The two gas analysers were arranged in series so that the same gas sample was monitored by both instruments. Gas was sampled at a flow rate of 0.5 l/ min., set by an adjustable pump on the carbon dioxide analyser. The electrical output of the CO_2 analyser was recorded on two channels of the Mingograf recorder. One channel recorded CO_2 concentrations in the 0-6% range; the second channel was fitted with a zero suppression unit and recorded CO_2 concentrations in the 6-14% range.

The oxygen analyser was also fitted with a zero suppression unit and recorded oxygen concentrations in the range 14-21% on the Mingograf recorder.

CALIBRATION OF THE ANALYSERS: The analysers were calibrated with four gas mixtures and room air. The calibration mixtures were contained in gas cylinders which had been analysed using a Scholander chemical gas analyser (Scholander, 1947). Each cylinder was analysed several times and an analysis was acceptable only when duplicate readings were within 0.05% of each other. The mixtures were used to prepare

calibration curves for the oxygen and carbon dioxide recording channels. Each curve was drawn from the readings of at least three gas mixtures which spanned the range of the recording channel.

The effect of high oxygen concentrations on the infra-red analyser reading of CO_2 (collision-broadening effect) was assessed experimentally; an increase of 10% in the oxygen concentration of a sample depressed the reading of a fixed concentration of carbon dioxide by 0.07%. Since the rebreathing procedure generated mixtures of CO_2 in approximately 50% oxygen, a maximum error of about 2 mm.Hg. therefore existed in the measurement of carbon dioxide during the rebreathing period. This degree of error in the measurement of mixed venous PCO_2 was considered negligible in view of the fact that the veno-arterial PCO_2 difference during exercise was usually some 30-40 mm.Hg. Moreover, this error would not affect the reproducibility of the method, or the comparison of different methods using high bag O_2 concentrations.

ELEMA-SCHONANDER MINGOGRAF-81 RECORDER: An 8-channel ink-jet recorder was used to record ECG, inspired and expired gas volumes and the concentrations of carbon dioxide and oxygen. The recorder was a fast-response instrument with a linear frequency response from D.C. to 500 Hz. and an amplitude linearity within $\pm$ 2%.

THE EXERCISE TEST

The experimental procedures used in these studies were the same, both at rest and during exercise.

Two exercise patterns were used in these studies. In one, a fixed work load was maintained for a prolonged period; serial measurements were then made at five minute intervals.

In the second type of test the period of rest or exercise (at a constant work load) was maintained for only a single five minute cycle of measurements, after which the work load was raised.

During the tests the subject breathed continuously from the valve-box assembly, with simultaneous measurement of electrocardiogram. An initial period of about 4 minutes was allowed for the subject to reach a steady state of heart rate, ventilation and gas exchange. This was assessed from an inspection of the recorded trace of the ECG, inspired gas volume and the mixed expired gas concentrations. When the steady state was achieved, expired gas was collected in the Tissot spirometer for a period of one minute for subsequent analysis of the mixed expired O_2 and CO_2 concentration; during the collection end-tidal PCO_2 was monitored continuously at the mouth. When the gas collection was completed, the subject rebreathed from an anaesthetic bag containing about 3 litres of a carbon dioxide in oxygen mixture. Rebreathing was continued for a period of about 15 seconds, during which the alveolar PCO_2 was monitored continuously for subsequent estimation of mixed venous PCO_2 (see below). At the end of the rebreathing procedure the steady state measurements were repeated either at the same work load, or at a new work load. The time required for each cycle of measurements was about 5-6 minutes.

MEASUREMENT OF MIXED VENOUS PCO_2

Two methods of measuring mixed venous PCO_2 ($P\bar{v}CO_2$) were used in this thesis. The plateau rebreathing method (Collier, 1956) was used to estimate $P\bar{v}CO_2$ from a plateau alveolar PCO_2 occurring in the course of a 10-12 second rebreathing procedure. The exponential rebreathing

procedure (Defares, 1958) estimated $P\bar{v}CO_2$ from the exponential change of alveolar PCO_2 as it approached the PCO_2 of mixed venous blood.

THE PLATEAU REBREATHING PROCEDURE: In the plateau rebreathing procedure, a rebreathing bag was filled with a mixture of carbon dioxide in oxygen which could equilibrate with the carbon dioxide contained in mixed venous blood prior to recirculation.

During the rebreathing period, the alveolar PCO_2 underwent three characteristic phases. There was an initial period of mixing between the bag and lung CO_2, which was indicated by a fluctuation of the alveolar PCO_2 monitored at the mouth in the first six seconds (Figure 2.2). Provided the initial level of CO_2 in the rebreathing mixture was correct, the release of CO_2 from venous blood was stopped completely within six seconds. Thus between 6-10 seconds there was a plateau in the alveolar PCO_2 since the CO_2 concentrations in the bag, the lung and mixed venous blood were in equilibrium. After ten seconds circulating venous blood re-entered the lung with a raised level of CO_2 and caused a steady rise in the alveolar PCO_2. The alveolar PCO_2 during the equilibrium period of 6-10 seconds was taken as the mixed venous PCO_2.

THE DENISON EXTRAPOLATION PROCEDURE: If the initial rebreathing bag mixture was inappropriate to the achievement of an equilibrium, a plateau could not occur within the given time limits; in this case the alveolar PCO_2 would continue to slope, due to a gradient of CO_2 gas tensions between mixed venous blood and the lung-bag system.

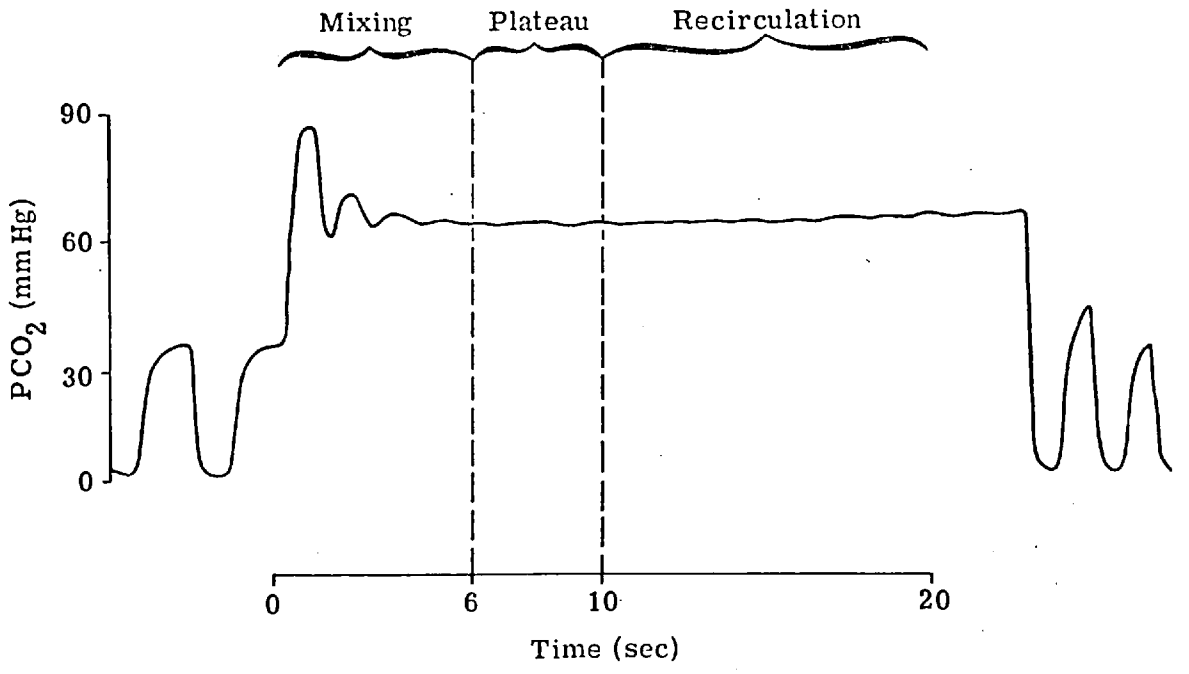


Figure 2.2 An infra-red CO_2 analyser record of PCO_2 during a plateau $P\bar{v}CO_2$ rebreathing manouvre (taken from Godfrey, 1974). Initial fluctuations in the record indicate mixing of CO_2 in the bag, lungs and blood. This is followed by an equilibrium (plateau) between the PCO_2 of mixed venous blood and the rebreathing volume. The rise in PCO_2 after the plateau is due to recirculation.

Denison (1968) and Edwards (1969) found that in such circumstances two procedures could be adopted; either the rebreathe could be repeated after a 1-2 minute interval, or the mixed venous PCO_2 could be estimated by a linear extrapolation of the sloping alveolar CO_2 concentration in the first rebreathe. The slope of the mean alveolar PCO_2 between the 6th and 10th second of the rebreathe was linearly extrapolated to a hypothetical $P\bar{v}CO_2$ measured at 30 seconds (at rest) or 20 seconds (during exercise) after the start of the rebreathe (Figure 2.3, taken from Godfrey, 1974). The alveolar PCO_2 measured at this time was then taken as the mixed venous PCO_2. Despite validation of the extrapolation procedure (Denison, 1968; Kelman, 1973) the reason for its validity remains obscure.

THE EXPONENTIAL REBREATHING PROCEDURE: In this method the subject rebreathed a mixture of 4-6% carbon dioxide in oxygen (Defares, 1958). In the present studies a range of rebreathing mixtures containing between 0-18% CO_2 in oxygen were investigated. The mixed venous PCO_2 was estimated from the exponential change in the alveolar PCO_2 during the rebreathing period. The $P\bar{v}CO_2$ was estimated through five different graphical solutions used by Klausen (1965), Defares (1958; 1972) and Jernerus et al. (1963).

1. THE GRAPHICAL SOLUTION OF KLAUSEN (1965): The end-tidal alveolar PCO_2 during the first fifteen seconds of the rebreathe (prior to recirculation) was plotted against time. Omitting the first breath, a smooth curve was drawn by eye through all subsequent points (Figure 2.4). The PCO_2 at one second intervals was noted;

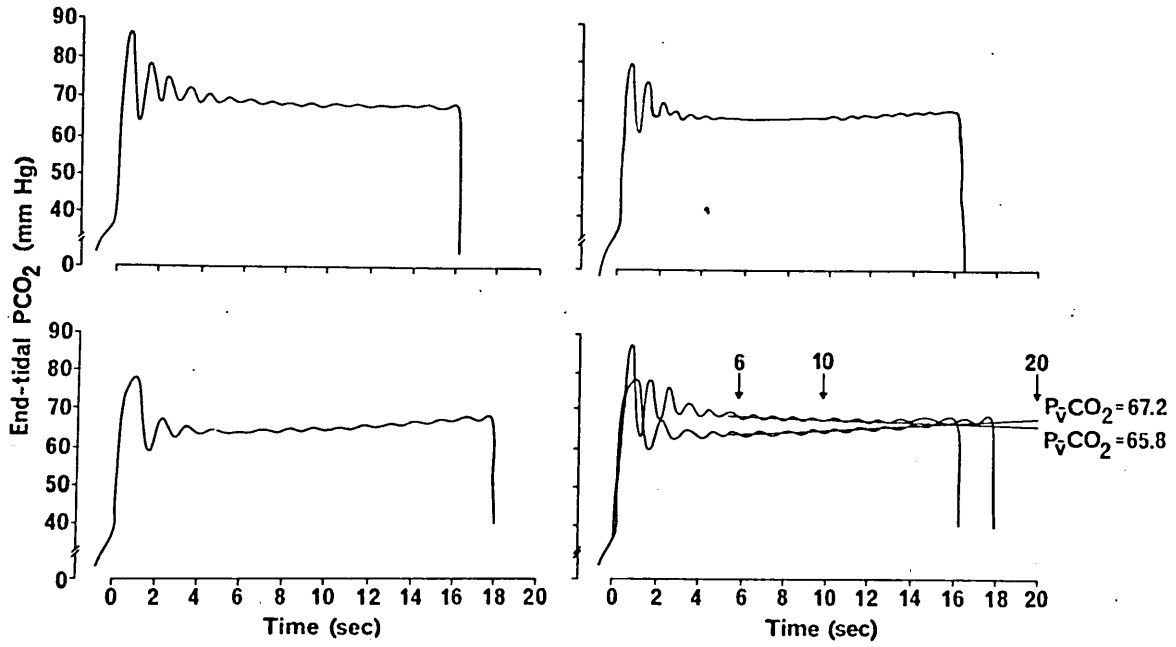


Figure 2.3 The Denison (linear) extrapolation procedure for the estimation of $P\bar{v}CO_2$ in the absence of a plateau (taken from Godfrey, 1974). In three consecutive rebreathing manouvres the initial CO_2 content of the rebreathing bag was varied. The initial CO_2 concentration in the first rebreathe was too high (top left), while that in the final rebreathe was too low (bottom left). An extension of the CO_2 slopes between the 6th and 10th second to the 20th second gives values of $P\bar{v}CO_2$ which are very close (bottom right) to the plateau estimate of 66mm. Hg. (top right).

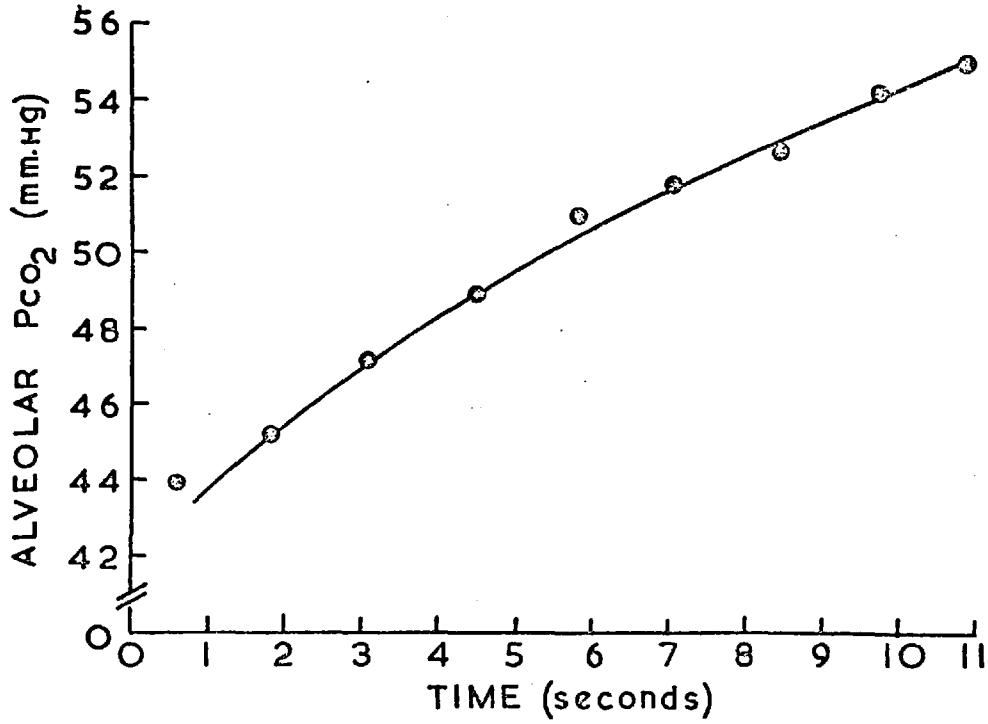


Figure 2.4 The exponential rise in alveolar PCO_2 during a Defares (exponential) rebreathing procedure, plotted against rebreathing time by the method of Klausen (1965). Ignoring the first breath the slope of the line is drawn by eye for all subsequent points.

the PCO_2 at each second (t_2) was then plotted against the PCO_2 at the previous second (t_1) for all values of PCO_2. A regression line was then drawn to intersect a line of identity where the PCO_2 at two consecutive times would theoretically be the same. This situation could only occur if the CO_2 tensions in the lung, bag and mixed venous blood were to remain unchanged over a period of time, that is, when they were in equilibrium. The PCO_2 at the point of intersection was therefore taken as the mixed venous PCO_2. (Figure 2.5).

2. THE GRAPHICAL SOLUTION OF JERNERUS et al. (1963): This method was similar to the second half of the Klausen method, but the initial tension: time curve (Figure 2.4) was omitted. The alveolar PCO_2s of consecutive breaths during the rebreathing period were simply plotted against each other to intersect a line of identity (Figure 2.6) which gave the value of $\bar{P}vCO_2$ as before.

3. THE GRAPHICAL SOLUTION OF KLAUSEN (using a modification by Defares): In this method a tension:time curve was initially prepared, as in Figure 2.4. For clarity, however, the difference in the alveolar PCO_2 at one second time intervals ($t_2 - t_1$) was plotted against the PCO_2 at time t_1. The difference in the PCO_2 at two consecutive times would theoretically be zero (PCO_2 being unchanged) only when equilibrium occurred, (see the Klausen method). The PCO_2 at the intercept, when the PCO_2 difference was zero, was therefore taken as the mixed venous PCO_2 (Figure 2.7).

4. THE GRAPHICAL SOLUTION OF JERNERUS (using a modification by Defares): This method simply plotted the difference in the alveolar PCO_2 of consecutive breaths against the PCO_2 of the initial breath. The

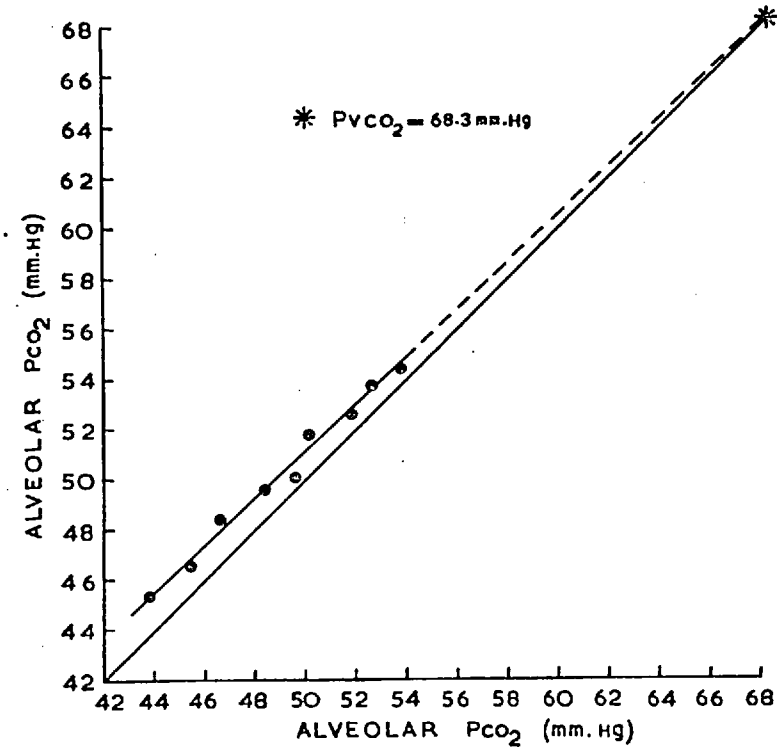


Figure 2.5 Mixed venous PCO_2 measured by the method of Klausen (1965). The alveolar PCO_2 at one second intervals (recorded from figure 2.4) is plotted against the alveolar PCO_2 of the preceding second. A line through all points is drawn to intersect a line of identity at which the alveolar PCO_2 of two breaths remains unchanged. The PCO_2 at the point of intersection measures the $P\bar{v}CO_2$.

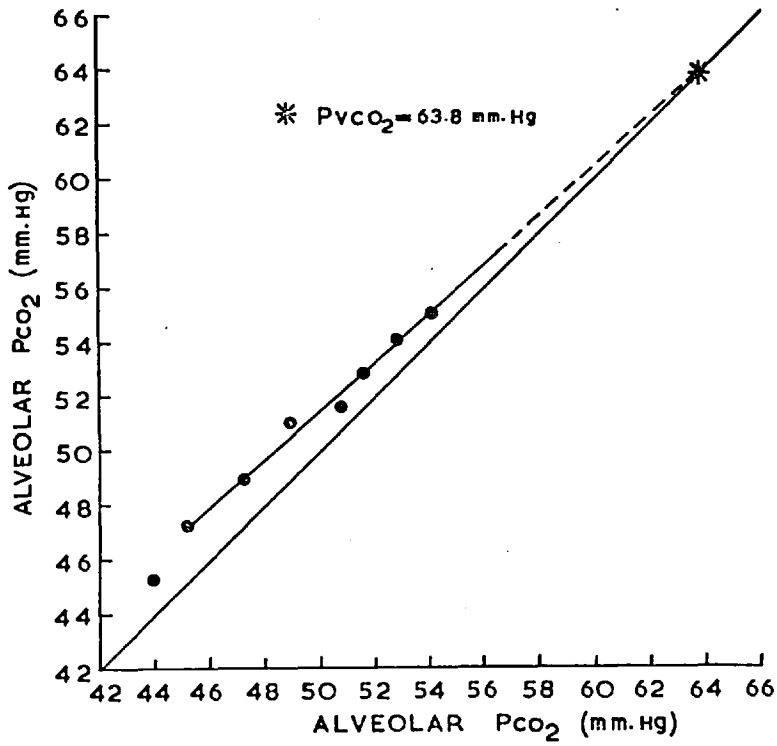


Figure 2.6 Mixed venous PCO_2 measured by the method of Jernerus et al. (1963). During a 10 sec. exponential rebreathing procedure the alveolar PCO_2 of one breath is plotted against that of the previous breath. Omitting the first breath, a line through all points is extrapolated to intersect a line of identity. The PCO_2 at the intersect is read as the $P\bar{v}CO_2$.

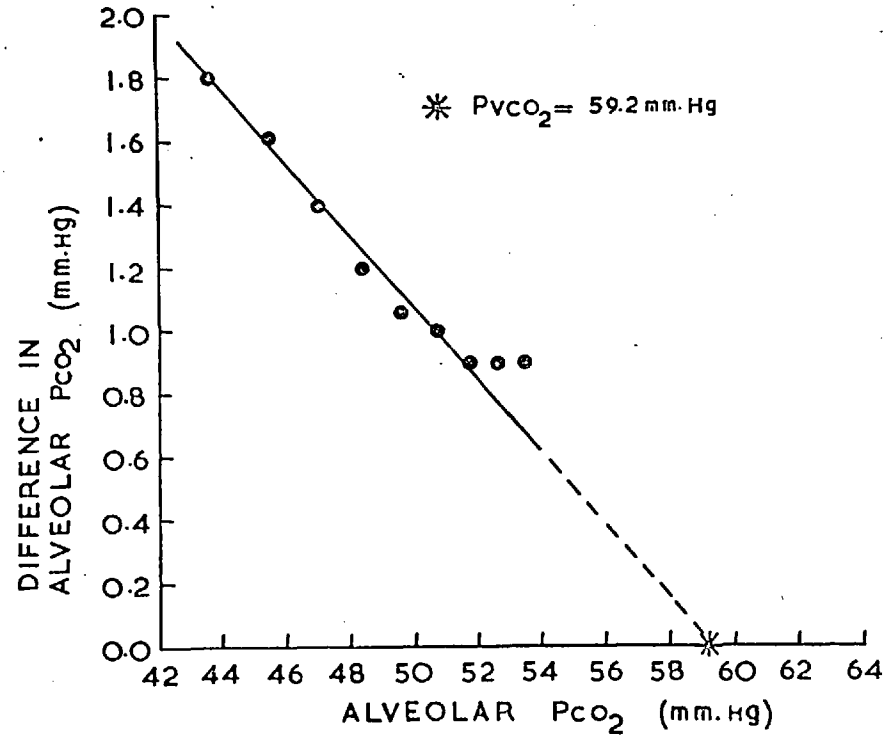


Figure 2.7 Mixed venous PCO_2 measured with a modification devised by Defares (1972). The difference in the alveolar PCO_2 at two consecutive times during a 10 sec. rebreathing procedure (measured as seconds or breaths), is plotted against the alveolar PCO_2 of the 1st value. A line through the points intercepts a value of alveolar PCO_2 at which there is no difference in the PCO_2 between samples. The PCO_2 at the intercept is taken as the alveolar PCO_2.

graphical solution would appear similar to the one shown in Figure 2.7.

5. THE GRAPHICAL SOLUTION OF DEFARES (1972): Defares (1972) suggested that an initial tension-time curve by the method of Klausen (1963) gave an unduly low value of $P\bar{v}CO_2$. He therefore suggested a new graphical solution for $P\bar{v}CO_2$. In this method the alveolar PCO_2 of consecutive breaths during the rebreathing period were noted. The difference in the PCO_2 between breaths (b_1 and b_2) was then calculated. This difference (ΔPCO_2) was corrected for the time interval of the two breaths ($\Delta PCO_2/\Delta t$) to give the rate of change of PCO_2 (mm.Hg./sec.). This value was then plotted against the PCO_2 of the initial breath (b_1). The value of PCO_2 when the rate of change of PCO_2 was zero was taken as the mixed venous PCO_2. Once again the graphical solution is similar to that shown in Figure 2.7.

ANALYSIS OF RESULTS AND CALCULATION OF CARDIAC OUTPUT

The results were calculated using a computer program (Godfrey, 1970) which processed and analysed all data. The program calculated each subject's gas exchange, ventilation and heart rate during the exercise test. In addition, the values of mixed venous PCO_2 were used to estimate cardiac output by the indirect (CO_2) Fick equation.

The calculations of ventilation, gas exchange and heart rate were carried out by standard arithmetic procedures. The cardiac output was calculated from the CO_2 production, divided by the veno-arterial content difference for CO_2 in blood.

THE CO_2 CONTENT OF BLOOD: The arterial and venous CO_2 contents of blood were calculated from the measured or assumed CO_2 gas tensions, using the carbon dioxide dissociation curve.

(1) The arterial PCO_2 was calculated from an estimate of the normal physiological dead space in each subject (Godfrey et al., 1970). The Bohr equation was used to estimate arterial PCO_2:

$$PaCO_2 = \frac{PECO_2}{(1 - \frac{VD}{VT})}$$

where VD = normal physiological dead space

VT = tidal volume

$PaCO_2$ = arterial PCO_2

$PECO_2$ = mixed expired PCO_2

(2) The arterial PCO_2 was used to calculate the arterial plasma CO_2 content, using a simple formula derived from the Henderson-Hasselbalch equation (McHardy, 1967).

(3) The arterial "total" blood CO_2 content was calculated from the plasma CO_2, allowing for the haemoglobin, pH and oxygen saturation of the blood. (McHardy, 1967; Godfrey, 1970).

(4) Before calculation of the veno-arterial CO_2 content difference, the slope of the logarithmic version of the CO_2 dissociation was defined for each subject. This slope was modified by the base excess of the individual's blood (calculated from the assumed or actual blood pH and bicarbonate levels) and its haemoglobin (Godfrey, 1970).

(5) The veno-arterial CO_2 content difference was then calculated from the slope of the CO_2 dissociation curve. The arterial PCO_2 and hence CO_2 content gave a fixed location on the dissociation curve, from which the venous CO_2 content ($C\bar{v}CO_2$) was estimated for a given value of the mixed venous PCO_2. The veno-arterial CO_2 content difference was then calculated with only a minor correction for the true oxygen saturation of arterial blood. (McHardy, 1967; Godfrey, 1970).

(6) Since both the carbon dioxide output and the veno-arterial content difference for CO_2 were then known, the Fick equation was used to calculate cardiac output.

CHAPTER 2

(b) THE NITROUS OXIDE REBREATHING METHOD

The apparatus and techniques for measuring pulmonary blood flow with a nitrous oxide rebreathing method were evolved over a period of time. In this period the theoretical and practical assumptions underlying the use of these methods were analysed. To limit the errors of some of these assumptions it was necessary to change parts of the apparatus and techniques.

Two experimental circuits were used; the first was similar to the one described for the CO_2 rebreathing method with minor modifications for the measurement of nitrous oxide. A fast response infra-red nitrous oxide analyser and a gas chromatograph were used to analyse gas concentrations of nitrous oxide and neon during an N2O-rebreathing procedure. A later section of this chapter describes a modified experimental circuit using a mass spectrometer in conjunction with an on-line computer for analysis of the data. The first nitrous oxide experimental circuit is described here.

The apparatus which was used to measure heart rate, ventilation and gas exchange in our subjects has been described in Section (a): The CO_2 rebreathing method. This section deals with additional units used for the nitrous oxide rebreathing experiments.

All the experiments were performed during steady state exercise at a fixed work load. The subject was seated on a bicycle ergometer and breathed through a rotary tap and valve-box assembly. Initial measurements of heart rate, ventilation and gas exchange were made in the first 4-5 minutes of the exercise test. The rotary plate assembly was then turned to connect the nitrous oxide rebreathing circuit to the subject.

A diagram of the nitrous oxide rebreathing circuit is shown in Figure 2.8.

The subject breathed through a second low resistance 3 way tap (5). A 6 litre open-ended anaesthetic bag (3) was attached to the distal end of the tap; by turning the tap, the subject could breathe either room air, or a nitrous oxide mixture from the rebreathing bag.

Small ports were drilled distally and proximally to the tap key. The distal port furthest from the mouth was used for continuous measurement of the nitrous oxide concentration in the rebreathing bag before the rebreathing procedure. The proximal port was close to the subject's mouth and was used to monitor the tidal concentration of the nitrous oxide during the rebreathing period.

A 3 way plastic tap (Pharmaseal K75) was turned to connect one of the two sampling lines to the fast response infra red nitrous oxide analyser (1 : Grubb-Parsons Med-1). An airtight pump (Charles Austen Ltd.) placed downstream

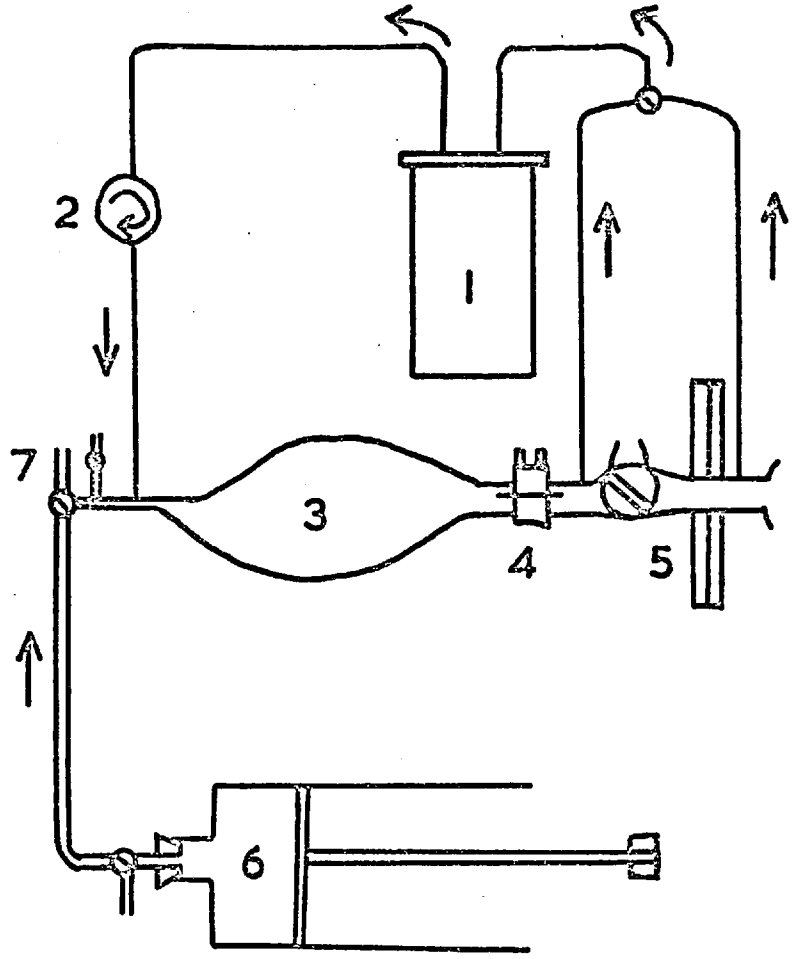


Figure 2.8 Diagrammatic representation of the N_2O rebreathing apparatus. (1) infra-red N_2O analyser (2) air-tight pump generating continuous flow of sampled gas (3) rebreathing bag (4) Fleisch pneumotachograph for measurement of air flow (5) 3-way tap and rotary turret for the introduction of air or rebreathing mixture to the subject (6) Gas mixing cylinder used to prepare rebreathing mixtures (7) sampling taps for the introduction or removal of gas mixtures.

(2) of the analyser was used to sample gas at a rate of 960 ml/min.

THE REBREATHING PROCEDURE: The nitrous oxide (N_2O) rebreathing manoeuvre was performed at intervals of five minutes during the exercise test. The 6 litre bag was initially evacuated using a suction line connected to a 3 way glass tap (7). A 4 litre mixture of 8% N_2O, 0.05% neon in air was then injected into the bag from a 5 litre perspex syringe (6). The N_2O analyser was used to monitor the initial bag concentration of N_2O. Using the distal sampling line, gas was passed from the bag, through the analyser and back to the rebreathing bag. In addition, a 40 ml sample of the bag mixture was removed into a 50 ml greased glass syringe for analysis of its neon concentration on a gas chromatograph (Carle Instruments 8000). A rebreathing procedure was then carried out. The subject breathed out to residual volume after which the main tap (5) was turned to connect the bag to the subject. The N_2O concentration at the mouth was monitored continuously from the proximal port throughout the rebreathing period. The distal sampling port was not used during the rebreathe, because of the large dead space of the tap (5) which was interposed between the sampling port and the mouth. The subject breathed deeply and rapidly (about 1 breath/second) for a period of about 10 seconds at the end of which he breathed out to residual volume once more. The tap (5) was then turned off and the distal sampling port was reconnected to sample the final mixed expired N_2O concentration in the bag. A second 40 ml. sample was withdrawn from the bag for estimation of the final neon concentration.

In the initial experiments a pneumotachograph (Fleisch No. 4) was used to monitor the phase of respiration during the rebreathing period (Ayotte et al. 1970). This was used to correct for a small time lag in the N_2O analyser readings which was due to the response-time of the instrument, and to dead space washout of the lung. Once the lag time of the instrument was determined the pneumotachograph was removed, and a correction was applied to all subsequent traces. The errors due to differences in the dead space washout time between subjects was minimal, especially after the first few breaths when the distinction between inspired and expired N_2O concentrations was extremely small.

DETAILS OF THE APPARATUS AND CALIBRATION PROCEDURES

The Infra-red Nitrous Oxide analyser (Grubb Parsons Med-1): The analyser was a fast-response instrument with an indium-antimonide split beam detector. The response time of the instrument was 0.1 seconds for 95% F.S.D. (nominal). The instrument was used to sample concentrations of N_2O through a 30 cm catheter tube (Portex 2/480). The pressure gradients from the analyser to all sampling ports were identical (the instrument was supposedly insensitive to flow rates below 1 l/min). Within the circuit the response time of the instrument was about 0.25 seconds. The instrument was insensitive to water vapour pressure and to carbon dioxide in concentrations well above 50%.

CALIBRATION OF THE INSTRUMENTS: The instrument was set to measure nitrous oxide concentrations in the range 0-9% N_2O. The sensitivity of the instrument was controlled by means of three potentiometers; which were adjusted to provide an almost

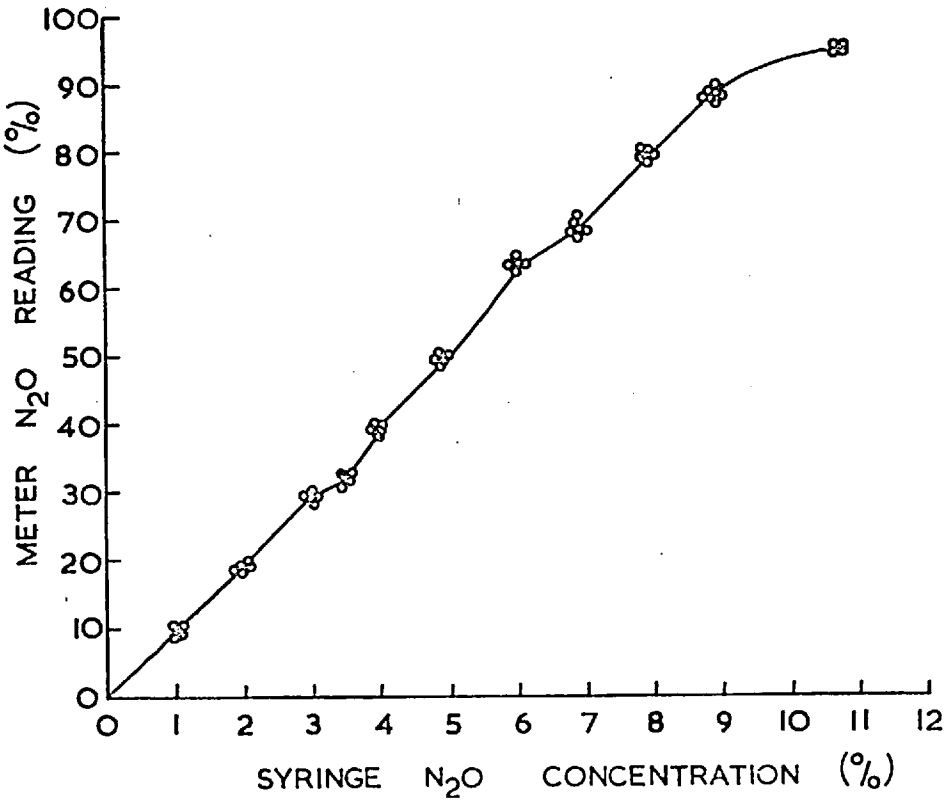


Figure 2.9 Calibration of the infra-red N_2O analyser meter readings against a range of N_2O concentrations prepared in a glass syringe. Alinearities in the readings of 3.5, 6.0 and 9.0% N_2O occur at the extremes in the sensitivity range of three potentiometers.

linear recording response; however at the extremes of each potentiometer's range (3.5%. 6.0%, 9.0% N_2O) the output signals were slightly warped (Figure 2.9). The distortions were not a cause of significant error in these studies since the initial and final N_2O concentrations in the rebreathing mixture were within the linearly recorded range. After each study the linearity of the analyser and recorder were checked several times with a range of gas calibration mixtures prepared in a gas syringe.

The signal from the analyser was recorded on two channels of a Mingograph 81 channel recorder. The first channel was used to record nitrous oxide in the range 0-9% N_2O. The second channel recorded a zero-suppressed signal in the range 4-8% N_2O (Figure 2.10) which was used to measure the alveolar N_2O concentration to an accuracy of 0.02%. Due to a mechanical beam chopper in the N_2O analyser the recorded signal had a slight sinusoidal interference pattern which did not interfere with the reading (Figure 2.10).

GLASS GAS CALIBRATION SYRINGE: A 100 ml glass syringe was used for the preparation of nitrous oxide calibrating mixtures; the syringe was initially calibrated by measuring the increase in weight due to the addition of a known volume of water. The results of the calibration are shown in Table 2.1. The syringe was subsequently used to calibrate the N_2O analyser with a range of N_2O gas mixtures. The gas concentration of each mixture was calculated from the markings of the syringe as corrected by Table 2.1.

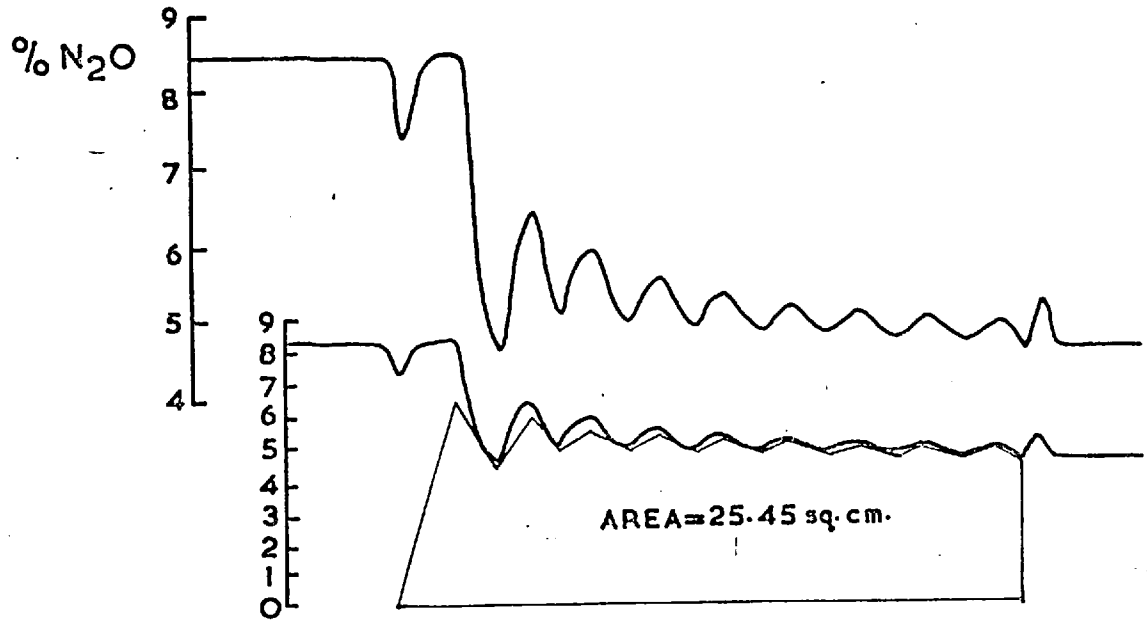


Figure 2.10 The alveolar N_2O concentration recorded during a 10 sec. rebreathing procedure, using an infra-red N_2O analyser and a Mingograf-81 recorder. The zero-suppressed upper signal was used to measure the end-expired alveolar N_2O concentration of each breath. The lower signal was used to calculate the time-integrated mean alveolar N_2O concentration after time delays in the instrument, and dead space washout in each subject had been allowed for.

Table 2.1: Calibration of the gas syringe by a water displacement method.

SYRINGE VOLUME READING (cc)	WEIGHT of WATER & SYRINGE (g)				SYRINGE VOLUME ACTUAL (cc)	
					$\bar{m}$	S.D.
00	354.60+	354.60+	354.60+	354.60+		
10	10.72	10.94	10.55	10.80	10.75 $\pm$	0.16
20	20.58	20.62	20.70	20.36	20.57	0.15
30	30.25	30.40	30.10	30.29	30.26	0.12
40	40.16	40.20	40.10	39.92	40.09	0.12
50	49.90	49.89	49.80	49.72	49.83	0.08
60	60.20	60.10	59.80	-	60.05	-
70	69.40	69.40	69.32	69.29	69.35	0.06
80	79.15	79.38	79.11	78.90	79.14	0.20
90	88.90	88.90	88.70	88.80	88.83	0.10
100	98.10	98.81	98.71	98.71	98.76	0.06

PERSPEX GAS SYRINGE: A 5 litre perspex syringe was used for the preparation of the 4 litre rebreathing mixtures. The volume markings of the syringe were calibrated against a water spirometer.

GAS CHROMATOGRAPH: A model 8000 Carle instruments gas chromatograph was used for the analysis of neon (and initally, nitrous oxide) concentrations in the rebreathing gas mixtures. Two ports on the instrument allowed the introduction of gas samples into two parallel sample columns. One column consisted of a six foot coiled stainless steel tube ($\frac{1}{8}$inch O.D.) containing a molecular seive. The other column contained Porapak Q 50/80, in a twelve foot length of tubing. Helium was used as the inert carrier gas which separated gas mixtures passing through the column. A dual chamber thermistor (100 μl volume) was used to detect the thermal conductivity of separate gas mixtures relating to the thermal conductivity of helium. The instrument was operated at a temperature of 70°C and a helium driving pressure of 15 p.s.i. Before the gas samples were introduced into the chromatograph columns they were stored in 50 ml glass syringes coated with silicone grease. The samples were flushed through two fixed-volume inlet ports on the chromatograph; the bulk of each sample was flushed through the system before its introduction into the helium gas stream of the chromatograph. Neon was separated and detected by the Porapak Q 50/80 column in about 7 minutes. In initial experiments, N_2O was also analysed chromatographically by the molecular seive column in 8 minutes.

TELSEC 700 CHART RECORDER: The output from the gas chromatograph was recorded on a flat-bed potentiometer chart recorder. The chart width of the recorder was 200 mm. for full scale deflection. In the operative range of voltage sensitivity the response time of the instrument was 0.3 seconds for full scale deflection.

CALIBRATION AND ACCURACY OF THE GAS CHROMATOGRAPH: Initially the gas chromatograph was used to measure the N_2O and neon concentrations in the gas mixtures. The absolute concentrations of N_2O and the relative concentrations of neon (measured as peak height in centimetres) were needed in the measurement of cardiac output. For four estimates of cardiac output it was necessary to analyse a total of 8 syringe samples for their concentration of neon and nitrous oxide; this required an analysis time of 56 and 64 minutes respectively. Since the gas samples were stored in 50 ml glass syringes (coated with silicone grease) it was necessary to know (a) whether this form of storage caused any change in the gas concentration of the samples over a period of 1-2 hours and (b) to establish the stability and accuracy of the chromatograph. A series of experiments were therefore carried out to assess these factors.

1. A gas mixture containing nitrous oxide and neon in air was stored in a sealed anaesthetic bag. Over a period of one hour a series of five 40 ml samples were withdrawn into a single 50 ml glass syringe. Each sample was immediately injected into the gas chromatograph. The results of one and two such runs (for

neon and nitrous oxide) are shown in Figure 2.11. There was some variation in the measurement of both nitrous oxide and neon, amountingto a maximum difference of 2% between readings. Assuming the gas concentrations in the bag remained unchanged over a one hour period, the 2% variation would imply an error in readings from the gas chromatograph.

2. Eight samples were withdrawn simultaneously from a bag mixture and were stored in 50 ml glass syringes. They were then analysed over a period of two hours. The results from four runs are shown in Figure 2.12 and indicate a variation of 3-5% in a single estimate of neon and nitrous oxide. The procedure adopted in this series of experiments was used in actual studies; however, since it was decided not to analyse N_2O on the chromatograph (see later) the total analysis time for neon alone would have been one hour. In this time period a 2% variation in the estimation of neon would be expected from Figure 2.12.

3. The linearity and stability of the chromatograph response to nitrous oxide and neon mixtures was examined. A series of 3 and 4 calibration curves were prepared at hourly intervals. The results are shown in Figure 2.13 (for nitrous oxide) and 2.14 (for neon). The response of the chromatograph was linear for the range of gas concentrations used in the studies; however, there was a drift in the slope of the calibration curve, which would have affected the absolute measurements of nitrous oxide and neon if a single calibration curve was assumed as true.

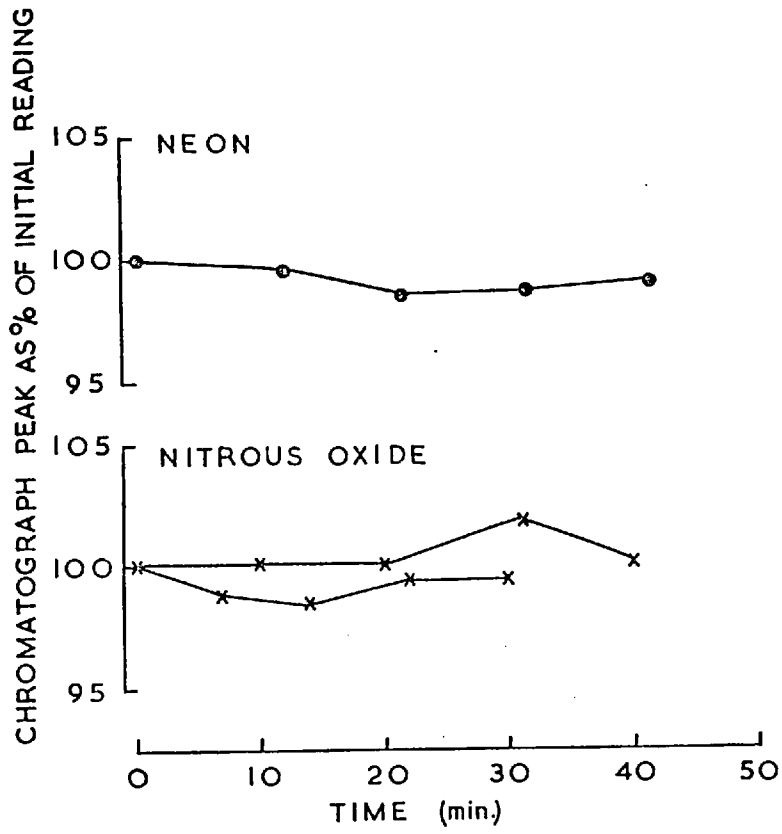


Figure 2.11 Variability of repeated Gas Chromatograph measurements of the neon and nitrous oxide concentrations of a gas mixture contained in a rebreathing bag for a prolonged period of time.

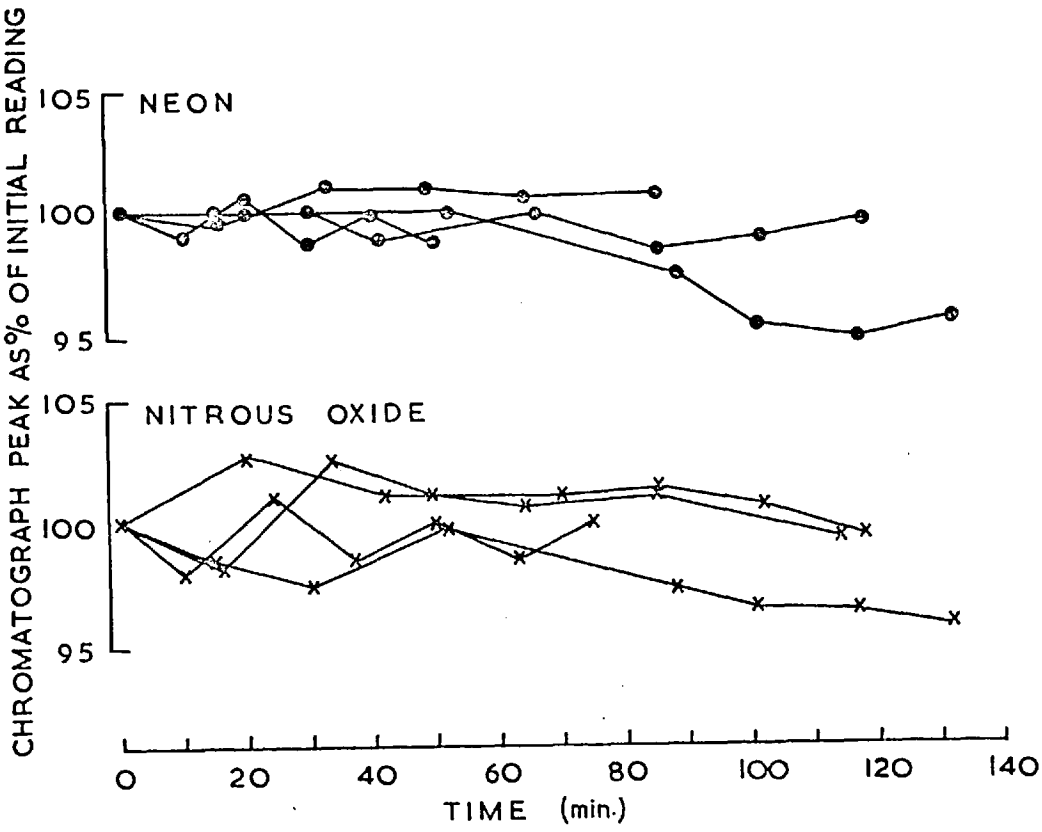


Figure 2.12 Variability of repeated Gas chromatograph measurements of the neon and nitrous oxide concentrations of an original gas mixture, stored in sealed glass syringes for measured periods of time.

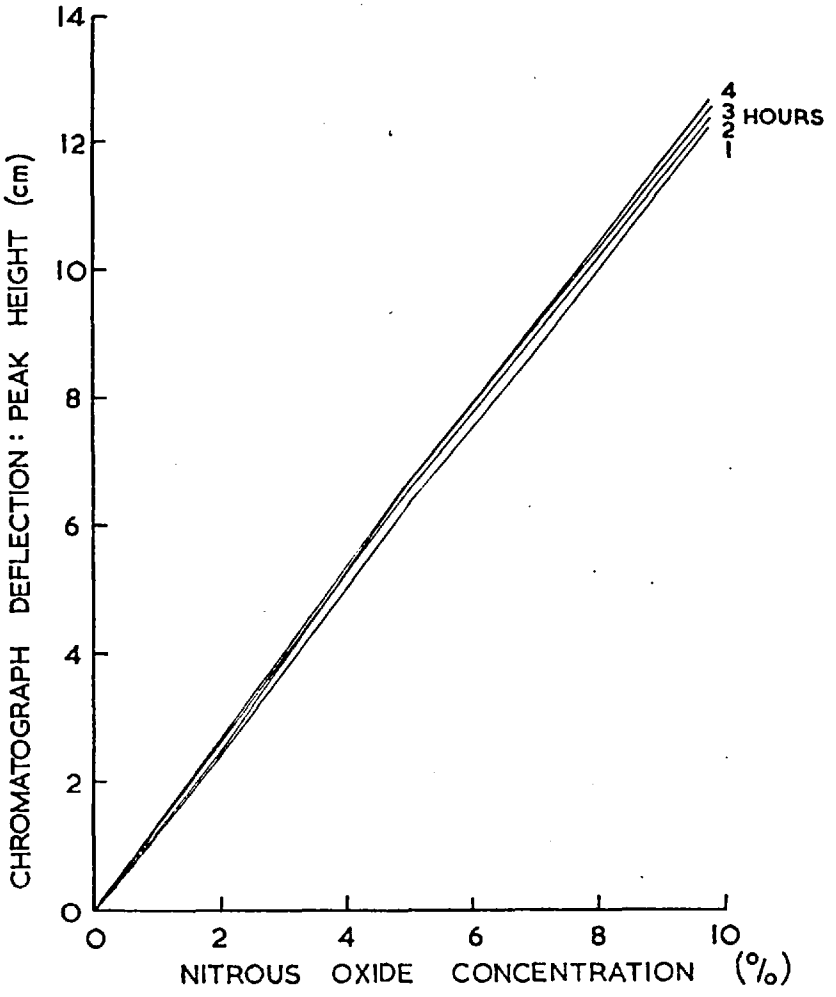


Figure 2.13 Calibration of the gas chromatograph for N_2O concentrations of less than 10%. Four calibration curves for N_2O were prepared at hourly intervals, in order to detect drifts in the sensitivity of the gas chromatograph. Calibrating mixtures were prepared in a 100 ml gas syringe.

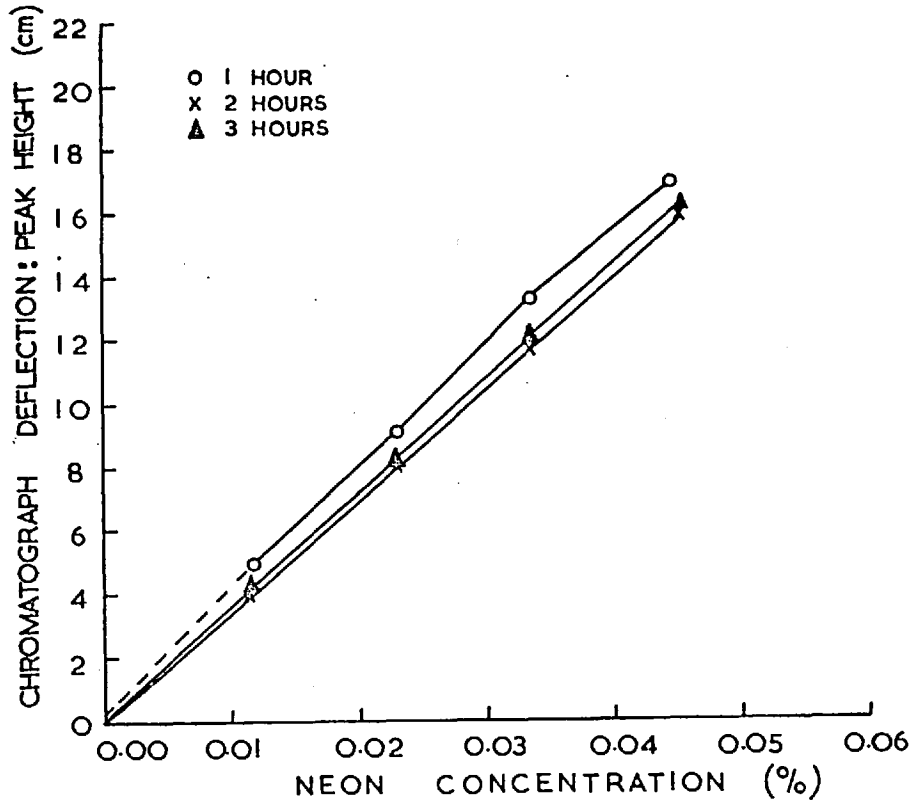


Figure 2.14 Calibration of the gas chromatograph for neon concentrations of less than 0.06%. The curves were prepared as described in figure 2.13, in order to detect changes in sensitivity of the gas chromatograph.

It was noted earlier that the proposed experiments required an absolute measurement of N_2O and only the relative peaks of neon. In view of this it was decided that in measuring N_2O the drift of the chromatograph was unacceptable for the method; N_2O concentrations were therefore measured on the infra-red N_2O analyser since this instrument was extremely stable after a warming up period of three hours (it was left on permanently). As before, the chromatograph was used to analyse the relative neon concentrations in the bag before and after the rebreathing procedure. The time interval for the analysis of the two samples from each rebreathe was seven minutes. In this time interval there would have been no significant drift in the calibration curve (Figure 2.14). The total analysis time for all the syringe samples was 49 minutes, during which period the concentration of neon in the syringe was not altered by more than $\pm$ 2% (Figure 2.12). In any case each pair of syringe samples was exposed to about the same period of "decay", so that the measurement of relative peaks was not significantly affected.

SOURCES OF TECHNICAL ERROR: Using the N_2O analyser, the concentration of nitrous oxide could be read to within 0.02% of a reading. This would have produced an error of $\pm$0.4 l/min in the estimate of cardiac output measured during exercise (Chapter 5). There was a $\pm$ 2% error in the estimation of neon, which was accumulated from the storage of syringe samples and drift of the chromatograph calibration. This would have produced an error of $\pm$0.4 l/min in the measurement of cardiac output. These errors were within the limits of error encountered by most methods of measuring cardiac output.

It should be noted that only technical errors have been discussed. The cumulative errors in the measurement of cardiac output are discussed later (Chapter 5).

ANALYSIS OF RESULTS

In both the Ayotte method (1970) and modified Cander and Forster method (1959), cardiac output ($\dot{Q}_C$) was calculated from the amount of nitrous oxide removed from the rebreathing bag by pulmonary blood ($\dot{V}N_2O$), divided by the mean alveolar concentration of N_2O during the rebreathing period ($F\bar{A}N_2O$) and the solubility of N_2O in blood (λB).

$$\dot{Q}_C \text{ (l/min)} = \frac{\dot{V}N_2O}{F\bar{A}N_2O \times \lambda B}$$

In describing the two methods it is perhaps useful to visualise the sequence of events during the rebreathing period. The bag is filled with a known volume of a gas mixture containing 10% N_2O. On inhalation of the mixture some N_2O is taken up instantaneously by lung tissue. During the rebreathing period the N_2O is mixed and diluted in the lung-bag volume. Within five seconds mixing is complete. However, throughout the ten-second rebreathing period there is a continuous (though declining) uptake of N_2O by pulmonary blood, which causes a diminution of the N_2O concentration in the bag. Concomitant with the drop in concentration, some of the gas initially dissolved in lung tissue is released into the alveolar gas phase. The amount of N_2O dissolved in lung tissue at the end of the rebreathe will then depend on the final N_2O concentration in the lung-bag system.

THE AYOTTE METHOD: The initial volume of the rebreathing bag (VB) was noted. The concentrations of nitrous oxide (FiN_2O) and inert gas neon (FiNe) in the bag were also recorded.

A ten-second rebreathing procedure was then performed, during which the alveolar N_2O concentration at the mouth was monitored continually. At the end of the rebreathing period the final concentrations of nitrous oxide and neon were recorded (FfN_2O, FfNe).

The amount of N_2O removed from the bag during the rebreathing period was calculated from the initial and final quantities in the bag and lungs.

1. The initial amount of N_2O in the bag was calculated from the bag volume (VB) and initial N_2O concentration (FiN_2O):

$$VB \times FiN_2O$$

2. At the end of the rebreathing period the N_2O was diluted in a lung-bag volume (VLB), estimated from the dilution of inert neon gas (FfNe).

$$VLB = VB \times \frac{FiNe}{FfNe}$$

3. The amount of N_2O in this volume was calculated from the concentration of N_2O remaining in the bag (FfN_2O):

$$VLB \times FfN_2O$$

4. The total amount of nitrous oxide removed from the system ($\dot{V}N_2O$) was therefore:

$$\dot{V}N_2O(1) = VB \times FiN_2O - VLB \times FfN_2O$$

5. However not all the nitrous oxide removed was dissolved in pulmonary blood. Some was contained in lung tissue. At the end of the rebreathe this amount (VTISS) was dependant on the final N_2O concentration in the system and the solubility of N_2O in lung tissue (λTISS). This amount was calculated as:

$$VTISS \times FfN_2O \times \lambda TISS$$

6. The nitrous oxide removed by pulmonary blood ($\dot{V}N_2O$) was therefore calculated from the total amount of N_2O removed during the rebreathe, allowing for solution of some N_2O in tissue:

$$\dot{V}N_2O = VB \times FiN_2O - FfN_2O\ (VLB + VTISS \times \lambda TISS)$$

7. The lung tissue volume (VTISS) was calculated indirectly from each subject's body weight, using the data of Sackner et al. (1964):

$$VTISS = 5.8\ (\pm 1.1) \times \text{body weight (kilograms)}$$

The lung tissue solubility constant for nitrous oxide (λTISS) was taken as 0.42 ml/ml of tissue/atmosphere (Ayotte et al. 1970).

8. The cardiac output was calculated by dividing the uptake of nitrous oxide by the mean alveolar concentration of nitrous oxide during the rebreathing period and the solubility of N_2O in blood (λB = 0.474 ml/ml/atmosphere; Kety, 1951). In the

Ayotte method the mean alveolar N_2O concentration was calculated from the area underneath the recorded rebreathing trace (Figure 2.10). The inspired and expired alveolar concentrations were determined after correcting for the response lag-time of the N_2O analyser. This was previously determined in several studies, in which a pneumotachograph was attached to the rebreathing bag.

During the rebreathing period the phase of respiration and the tidal concentration of N_2O were monitored simultaneously. The readings of the tidal N_2O concentration (measured on the N_2O analyser) could then be corrected against the true phase of breathing measured on the pneumotachograph.

The slopes of the alveolar concentration during each expiratory breath were extrapolated to the inspiratory phases of the breath before and after the expiration, to give the alveolar concentration of N_2O during the inspiratory phase.

The area which was measured under the corrected rebreathing trace (Figure 2.10) represented the product of the mean alveolar N_2O concentration and the rebreathing time (the time integral of the alveolar N_2O concentration) in units of atmosphere-seconds. Multiple planimeter readings of the area were accepted only when they agreed to within $\pm$ 1% of each other. A variation of $\pm$ 1% represented a possible error of $\pm$ 0.2 l/min in the measurement of cardiac output.

The final form of the Ayotte equation for calculating cardiac output ($\dot{Q}C$) was -

$$\dot{Q}C = \frac{\dot{V}N_2O}{\int_0^t FN_2O\,dt \times \lambda B}$$

where $\dot{V}N_2O$ = uptake of N_2O by pulmonary blood.

λB = solubility of N_2O in blood (0.474 ml/ml/atmosphere)

$\int_0^t FN_2O\,dt$ = time integrated mean alveolar N_2O concentration.

THE MODIFIED CANDER METHOD: The original breathholding method for measuring cardiac output (Cander and Forster, 1959) was modified to a rebreathing procedure.

As in the Ayotte method, the initial bag volume (VB) was known.

1. The total lung-bag volume (VLB) during the rebreathing period was calculated from the initial and final diluted concentrations of neon before and after a rebreathing procedure (FiNe, FfNe).

$$VLB = VB \times \frac{FiNe}{FfNe}$$

2. The lung tissue acted as a freely exchanging reservoir of dissolved nitrous oxide and could be considered as an equivalent volume of N_2O ($VTISS \times \lambda TISS$) which was added to the lung-bag volume. The total rebreathing volume (VTOT) was then comprised of:

$$VTOT = VLB + VTISS \times \lambda TISS$$

where as before, VTISS = 5.8 ($\pm$1.1) x body weight (kilograms)

$\lambda TISS$ = solubility constant of N_2O in lung tissue

(0.42 ml/ml/atmosphere)

3. The rate of uptake of nitrous oxide from the total rebreathing volume was estimated from a semilogarithmic plot of the end-expired N_2O concentration during the rebreathing pericd. An example is shown in Figure 2.15, which was prepared from the data in Figure 2.10.

In the first few seconds of the rebreathe there was a pericd of gas mixing between the lung and bag compartments (Figure 2.10). The end-expired N_2O concentrations in this period were therefore omitted from the semilogarithmic plot. After the mixing period, the exponential loss of N_2O from the rebreathing system (and its removal by pulmonary blood) was calculated from the regression slope of the end-expired N_2O concentration between 3-10 seconds during the rebreathing period.

4. The linearity of the semilogarithmic plot of N_2O reflects an exponential rate of uptake of N_2O by blood. The rate of uptake of N_2O at any time is therefore related to the concentration of N_2O in the lung-bag system at that time.

The exponential uptake is of the form:

$$(1) \qquad \frac{dFAN_2O}{dt} = FAN_2O \times k$$

where FAN_2O = the end-expired alveolar concentration of N_2O at a given time during the rebreathe.

5. It is obvious that the volume of N_2O in the system decreases because of its removal by pulmonary blood. This change will depend upon the rate of blood flow through the lungs ($\dot{Q}c$),

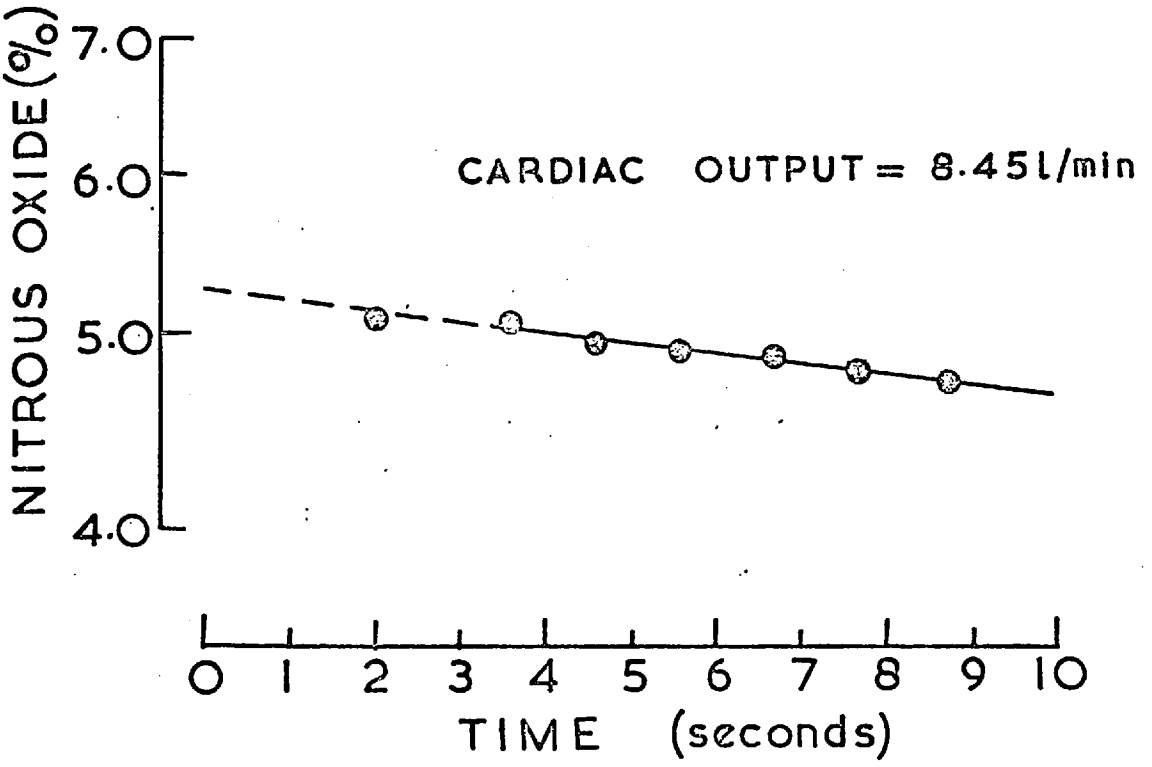


Figure 2.15 The end -expired alveolar N_2O concentration of each breath during a rebreathing period is plotted on semi-logarithmic ordinates against rebreathing time.

the solubility of N_2O in blood (λB) and the instantaneous concentration of nitrous oxide (FAN_2O) in the alveoli. The rate of loss of N_2O ($d\dot{V}N_2O/dt$) is therefore given by

$$(2) \qquad \frac{d\dot{V}N_2O}{dt} = FAN_2O \times \dot{Q}C \times \lambda B$$

Since, at any instant, the volume of N_2O (VN_2O) in the system is a product of the concentration (FAN_2O) and the volume in which it is diluted (VTOT), the rate of change in the N_2O concentration is given by:

$$(3) \qquad \frac{dFAN_2O}{dt} = FAN_2O \times \frac{\dot{Q}C \times \lambda B}{VTOT}$$

where VTOT, the total rebreathing volume remains constant.

Comparison of equation (3) with equation (1) indicates that -

$$(4) \qquad k = \frac{\dot{Q}C \times \lambda B}{VTOT}$$

Equation (1) can be integrated to give:

$$(5) \qquad FAN_2O_{t_2} = FAN_2O_{t_1} \times e^{-kt}$$

which can be rewritten as:

$$(6) \qquad FAN_2O_{t2} = FAN_2O_{t1} \times e^{-\frac{\dot{Q}C \times \lambda B}{VTOT} \times t}$$

where FAN_2Ot_2 = end-expired alveolar concentration of N_2O at time t_2.

FAN_2Ot_1 = end-expired alveolar concentration of N_2O at time t_1.

e = exponential function (e = 2.313)

t = The time interval $(t_2 - t_1)$.

Rearranging equation (6):

$$(7)\quad \frac{FAN_2Ot_2}{FAN_2Ot_1} = e^{-\frac{\dot{Q}C \times \lambda B \times t}{VTOT}}$$

Equation (7) is a mathematical statement of the fact that the loss of nitrous oxide from the lung-bag system is exponentially related to its removal by the pulmonary blood. The pulmonary blood flow ($\dot{Q}C$) can therefore be calculated by altering equation (7) to give:

$$(8)\quad \dot{Q}C = \frac{(VTOT)}{(\lambda B x t)} \times -\ln \frac{FAN_2Ot_1}{FAN_2Ot_2}$$

Alternatively the equation can be rewritten:

$$\dot{Q}C = \frac{(VTOT)}{(\lambda B x t)} \times \ln \frac{FAN_2Ot_2}{FAN_2Ot_1}$$

(c) THE MODIFIED NITROUS OXIDE REBREATHING METHOD

When the N_2O rebreathing methods of Ayotte, and of Cander and Forster (modified) were examined in Chapter 5, it became obvious that they both had inherent experimental errors which affected the calculation of cardiac output. In addition the experimental procedures were time-consuming and cumbersome. Several modifications were therefore carried out to improve the accuracy of the N_2O rebreathing method, and to increase the rapidity of all experimental measurements.

A mass-spectrometer was used in conjunction with an on-line computer to allow a breath-by-breath analysis of pulmonary gas exchange (Davies, et al. 1974); during a rebreathing procedure the end-tidal concentrations of several gases (including argon, CO_2 and N_2O) were also measured. In the previous N_2O rebreathing methods the total lung-bag volume was assumed to remain unchanged during the rebreathing period (section b of this chapter). This was found to be a source of serious error (Chapter 6), and in the present method changes in lung-bag volume during the rebreathing period were recorded from changes in the concentration of an inert gas, argon. The 5-channel mass-spectrometer allowed the simultaneous measurement of the N_2O and argon concentration during a rebreathing procedure. Unlike the previous methods, the analysis of all gas concentrations was virtually instantaneous.

Details of the equipment are shown in Figure 2.16. The subjects exercised on a bicycle ergometer in the upright sitting position. Inspired gas was inhaled through a dry gas meter which had been calibrated against a Tissot spirometer. A potentiometer signal from the gas meter was passed into an 8-channel recorder and then to a 4K capacity computer (Digico).

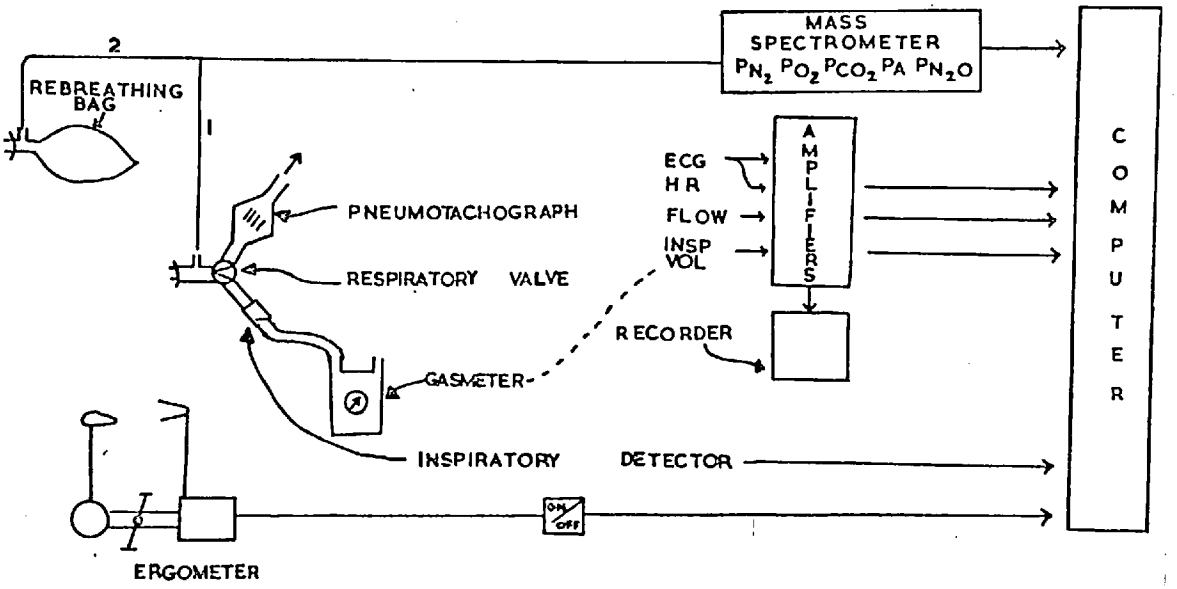


Figure 2.16 Diagrammatic representation of the apparatus used for exercise testing. The apparatus incorporated on-line computer facilities, and allowed the use of the modified N_2O rebreathing method.

The inspired air passed through an inspiratory detector which was comprised of a plastic hinged flap fitted with a silver contact. Elevation of the flap at each inspiration interrupted an electrical signal, which in turn triggered the computer measurement of inspiratory and expiratory gas concentrations (and flow rate) in the next breath.

The subject inspired air through the inspiratory port of a low-resistance valve with a 60 ml respiratory dead space. The resistance of the valve and circuitry are shown below:

	Flow Rate (l/min.)	Resistance (cmH_2O/l/sec.)
Inspiration	60	1.00
	120	0.75
Expiration	60	0.50
	120	1.00

Inspired and expired gas was sampled continuously at the mouth, using a modified mass spectrometer (MS4-AEI). The instrument analysed all gas mixtures for the fractional concentrations of N_2, O_2, A, CO_2 and N_2O. The output from the instrument was fed into the computer.

The flow rate of expired gas was measured through a heated pneumotachograph in conjunction with a pressure transducer. The signal was amplified and recorded by the computer and a chart recorder.

At timed intervals between "steady state" measurements of heart rate, gas exchange and ventilation, a rebreathing procedure was carried out to measure cardiac output by either the nitrous oxide or carbon dioxide gas rebreathing methods. The steady state was assessed from a constant heart rate and ventilation over the exercise period.

DETAILS OF THE APPARATUS

THE MASS SPECTROMETER (MODIFIED MS4-AEI): All gases were analysed on a 5-channel respiratory mass spectrometer which sampled inspired and expired gases at the mouth continuously. The five channels monitored the gas concentrations of N_2, O_2, A. CO_2 and N_2O. A sampling probe from the mass spectrometer was fitted tightly into the mouthpiece, 2-3 cm. from the subject's mouth. The probe (inlet diameter 0.025 cm.) sampled gas at a rate of 25 ml/minute. The instrument had a delay time of less than 100 milliseconds, which was noted by the computer in all calculations of gas exchange.

The design of the gas sampling system is shown in Figure 2.17 (taken from Davies, et al. 1974). Gas sampled at the tip of the probe (atmospheric pressure) was passed through a large pressure gradient along the length of the inlet tube. By the time the gas was analysed from Chamber A (through a molecular leak B), the pressure of the sample was only 5 mmHg. This system was susceptible to some drift instabilities due to three main causes:

1) Changes in the viscosity of gases could alter the pressure in Chamber A, which in turn would alter the flow of sampled gas. The changes in viscosity could be caused by a variation in the temperature or composition of the gases.

2) Partial blockage of the probe would also add to the instability of the mass spectrometer measurements by altering the partial pressure gradient of the sampling system.

3) Finally, with its slower rate of movement, water vapour could produce phase errors in the measurement of gas concentrations.

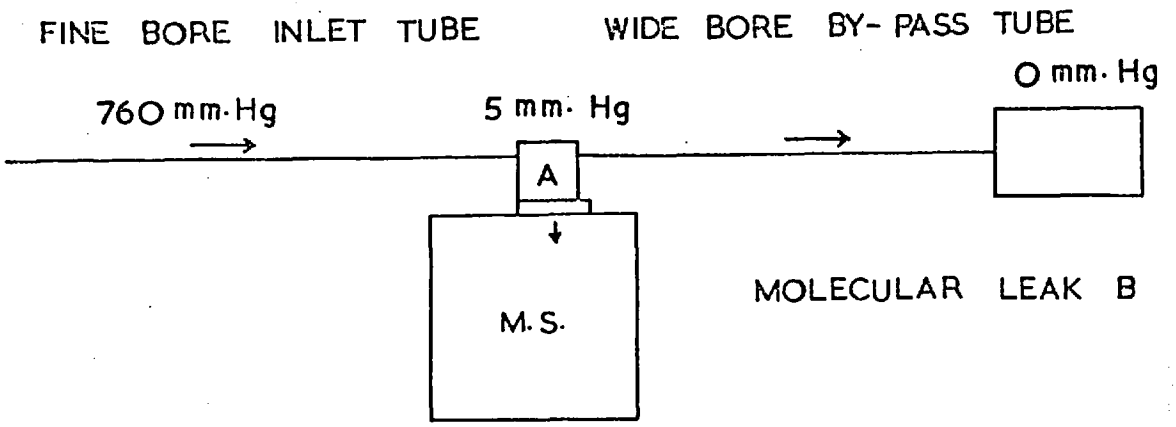


Figure 2.17 Block diagram of the sampling system of the mass spectrometer (taken from Davies et al, 1974). Gases sampled at atmospheric pressure undergo a pressure reduction to about 5mm.Hg. as they pass through a narrow inlet tube to chamber A. Most of the gas is released via a wide-bore by-pass tube; a small fraction enters the mass spectrometer's ion-source region through a molecular leak B, for subsequent detection.

The computer was used to compensate for these problems by constantly dividing individual partial pressures by the summed total of partial pressures; this procedure gave the dry gas fractional composition of each gas. With further refinements it allowed considerable accuracy in the measurement of gas partial pressures. Individual gas tensions measured on the mass spectrometer differed from the true reading by no more than 1% (e.g. 10% ± 1% of 10%). In addition the recorded tension could differ from the true tension by a further 0.1% due to a drift in the zero baseline of the instrument. (e.g. 5.0% ±0.1%). However, at any instant these errors would have affected all gases equally and were largely eliminated by the summation procedure on the computer.

THE FLEISCH (No. 4) PNEUMOTACHOGRAPH: Expiratory flow rates were measured with a pneumotachograph and an Elema Schonander pressure transducer. The flow sensitivity of the instrument was affected by changes in temperature, humidity and density of gases. The instrument was therefore maintained at a temperature of 37°C and the computer was used to adjust the calibration of the instrument continually against simultaneous measurements of the inspired volume from the gas meter. The procedure allowed a long-term calibration, on the assumption that there was no net absorption or excretion of nitrogen in the lungs during the exercise test (Davies, et al. 1974).

THE REBREATHING CIRCUIT: A 10 litre anaesthetic bag was filled with a 4 litre mixture of either:

a) carbon dioxide in 50% oxygen, remainder nitrogen

or b) 10% nitrous oxide in 25% oxygen, 3-4% Argon, remainder nitrogen.

The gas mixtures were initially prepared in high pressure gas cylinders and were then used as required. The rebreathing bag was filled from a 5 litre perspex syringe (described earlier) and was connected to the subject by an automatic switching device. An electrical signal from the device produced a pne matic pulse to activate a plate shutter at the end of a normal expiration. The shutter closed off the normal breathing circuit and connected the bag to the subject at inspiration. During the rebreathing period a pressure manometer was used to measure the inspiratory and expiratory pressure fluctuations at the mouth, so that the computer could monitor the subject's rebreathing pattern. After a suitable re-breathing period the shutter automatically reconnected the valve-box assembly to the subject, who then resumed normal breathing.

OPERATION OF THE APPARATUS: An on-line computer program was used for the assimilation of all data. Before each test the values of barometric pressure, room temperature and humidity were fed into the computer. These calibrations allowed the computer to calculate gas volumes and concentrations in units of standard temperature and pressure (dry): S.T.P.D. The computer was then given a value for the bag gas volume which was used in the rebreathing procedure; this was subsequently used to calculate changes in the total lung-bag volume during the rebreathing procedure (in S.T.P.D. units). The mas spectrometer and computer were then calibrated with physiological concentrations of N_2, O_2, A, CO_2 and a mixture of 10.6% N_2O (in 50% oxygen and remainder N_2). The computer used the calibrations to calculate the dry gas fractions of all five gases. By giving the calibration gases singly and in combinations, the computer was able to

correct for cross-interference in the signal of gases having the same molecular weight. In the case of nitrous oxide (MW.44) there was considerable interference from carbon dioxide (MW44). The mass spectrometer was therefore set to read the peak concentration of nitric oxide, a breakdown product of nitrous oxide. A similar cross-interference between nitrogen and carbon monoxide (breakdown product of CO_2) was corrected by the computer.

A spot analysis of room air and other calibrating gases confirmed the accuracy of mass spectrometer and computer readings before actual experiments were performed (Table 2.2).

In each experiment the subject's ECG was recorded continuously. The QRS pulse of the ECG was also fed to the computer for the calculation of heart rate. The inspired gas volume and the expired gas flow rate during normal breathing were measured breath by breath; the gas concentrations of N_2, O_2, A, CO_2 and N_2O throughout the breathing cycle were measured simultaneously and these measurements were continued during the rebreathing period.

The information from each test was stored on punch-tape, for subsequent analysis by an off-line computer program.

ANALYSIS OF RESULTS

An off-line computer program was used to analyse the raw data assimilated on the punch tape. The program calculated values for heart rate, ventilation and gas exchange during each test. The alveolar concentrations of the five gases were calculated for each breath during the rebreathing period.

Table 2.2: Spot analysis of room air using the mass spectrometer linked to an on-line computer.

MULTIPLE SPOT ANALYSES of AIR.

DRY GAS FRACTIONAL CONCENTRATIONS of:

NITROGEN	OXYGEN	ARGON	CARBON DIOXIDE	NITROUS OXIDE
0.7800	0.2106	0.0094	0.0001	- 0.0001
0.7805	0.2101	0.0093	0.0001	- 0.0001
0.7797	0.2108	0.0094	0.0001	- 0.0001
0.7805	0.2101	0.0094	0.0001	- 0.0001
0.7802	0.2104	0.0093	0.0001	- 0.0001

The results of each test were presented by a teletype data output sheet (Table 2.3). The calibration data used in the tests were displayed first, under a code number [0] . The mean values of heart rate, ventilation and gas exchange at 20 second intervals during the test were then presented under a code number [3] . Additional data in [3] included the mixed-expired and end-tidal concentrations of oxygen and carbon dioxide.

If at any stage the measurements were interrupted by a rebreathing procedure, a third set of results were printed out under a code number [4] . Each column listed the mean inspired, the alveolar mean expired and the alveolar end-expired concentrations of N_2. O_2, A, CO_2 and N_2O in each breath during the rebreathing period. The data was used to calculate cardiac output by the CO_2 or N_2O rebreathing methods.

Cardiac output was measured from the uptake of nitrous oxide by pulmonary blood during a ten-second rebreathing procedure. The removal of nitrous oxide from the rebreathing system was corrected for the simultaneous changes in the lung-bag volume in this period. In this respect the method differed from the Ayotte method (Ayotte et al. 1970) and the Cander and Forster method (1959), and was similar to the methods of Rigatto (1967), Saltin (1964) and Pugh (1972).

Prior to any calculations the mean alveolar concentrations of nitrous oxide and argon during the rebreathing period (Code [4] , Table 2.3) were plotted against time. The N_2O and Argon concentrations were plotted on semilogarithmic and linear ordinates respectively (Figure 2.18). In the case of N_2O, the semilogarithmic plot was used linearise the slope of the exponential loss of nitrous oxide

Table 2.3: Format of the computer output data.

SWITCH CODE:

(0) INITIAL INPUT DATA

Reference No.	Body Wt (Kg)	P. Bar (mm Hg)	W.V.P. (mm Hg)	Room Temp. (^{o}C)
1	70	752	13	20

Gas meter sweep (l)	Cal Gas O_2(%)	Cal Gas CO_2(%)	Cal Gas N_2O(%)	Rebreathing bag volume (l)
10	16.20	5.40	10.60	4.000
				3.688 (l.S.T.P.D.)

(3) STEADY STATE MEASUREMENTS

Time (Sec.)	Heart rate (bts/min)	Breathing frequency (/min)	Tidal Volume (l)
22.7	116	26	1.159

Ventilation (l/min)	Oxygen consumption (l/min)	Carbon dioxide output (l/min)	Respiratory exchange ratio
30.6	1.180	0.920	0.78

Mixed expired oxygen (%)	Mixed expired carbon dioxide (%)	End-tidal oxygen (%)	End-tidal carbon dioxide (%)
14.15	3.70	12.70	4.78

(4) REBREATHING MEASUREMENTS

Time (Sec.)	Rebreathing time (Sec.)
186	1.36

Table 2.3: continued.

DRY-GAS FRACTIONAL CONCENTRATIONS of:					
N_2	O_2	A	CO_2	N_2O	
0.5882	0.2556	0.0528	0.0032	0.0999	mean inspired concentration
0.6847	0.1880	0.0318	0.0469	0.0483	mean expired alveolar conc.
0.7011	0.1758	0.0287	0.0529	0.0413	end-expired concentration

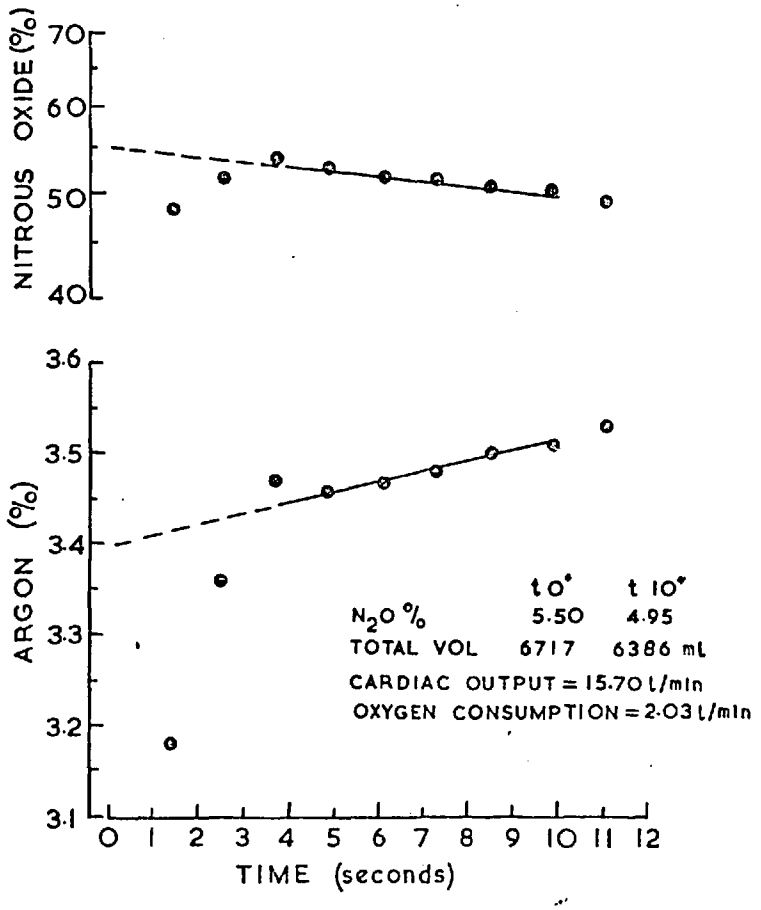


Figure 2.18 The end-expired alveolar concentrations of N_2O and Argon(measured breath by breath during a 10 sec. rebreathing procedure) are plotted on semi-logarithmic ordinates against rebreathing time. Initial fluctuation in the gas concentrations indicates a period of gas mixing. After 4 sec. mixing is completed, and is followed by a continuous loss of N_2O from the system; simultaneous shrinkage in rebreathing volume is monitored from the increase in argon concentration. Omitting the first few breaths, the slope of the concentration curve is used to calculate the net uptake of N_2O from the initial and final volumes of N_2O in the system. After a small correction, true N_2O uptake is used to calculate cardiac output by the Bornstein equation.

from the lung-bag system; the argon concentration was plotted on linear ordinates on the assumption (discussed later) that it's change was the product of an exponential loss of N_2O which was balanced by an exponential release of CO_2 into the lung-bag system, thus leaving a constant rate of removal of oxygen from the system.

1. The plot of argon indicated an initial mixing period of 4-5 seconds. After this period there was a linear increase in the argon concentration, due to a shrinkage in the lung-bag rebreathing volume. The shrinkage was due to an imbalance in the exchange of N_2O, O_2 and CO_2 across the lung (as discussed above), the net effect of which was a removal of gas from the lung-bag system. The volume of the rebreathing system was calculated from the argon concentration (see later).

2. The semi-logarithmic plot of nitrous oxide also showed an initial period of mixing, after which there was a continuous exponential loss of N_2O from the lung-bag system due to its uptake by pulmonary blood. Recirculation of venous blood after about 10 seconds caused a shallowing of the N_2O slope, since the back-pressure of N_2O in venous blood reduced uptake from the lung-bag system.

3. The slopes of the argon and nitrous oxide concentration between 4-10 seconds were used to calculate the uptake of nitrous oxide by pulmonary blood. To simplify the arithmetic calculations, the slopes were extrapolated backwards to zero time, so that measurements were standardised to a 10 second rebreathing period.

CALCULATION OF LUNG-BAG VOLUME. AND UPTAKE OF NITROUS OXIDE: The lung-bag volume at times zero (extrapolated) and ten seconds were calculated from the argon trace.

1. The initial volume of the rebreathing bag (VB) was known; the concentration of argon in the bag (A bag) was monitored from the first inspiratory breath of the rebreathing period. Before the rebreathe the argon concentration in the lung was assumed to be the same as that of room air.

2. During the rebreathe, the concentrations of argon in the bag and lung were fully mixed within four seconds, after which variation in the mean alveolar concentrations of argon reflected changes in the lung-bag volume (V_{LB}). The mixed expired concentrations of argon at an extrapolated time of zero (A_o), and at ten seconds (A_{10}) were calculated from the equations:

(i) V_{LB} at time zero seconds = $VB \times \frac{Abag - Ao}{Ao - Aroom} + VB$

(ii) V_{LB} at time ten seconds = $VB \times \frac{Abag - A_{10}}{A_{10} - Aroom} + VB$

3. It was noted earlier (see this chapter, section b) that a reservoir of N_2O was dissolved in lung tissue during the rebreathing period. A correction was made for this when pulmonary blood flow was calculated.

As before, the total volume of nitrous oxide was diluted in a total rebreathing volume (V_{TOT}), which was comprised of the lung-bag (V_{LB}) and equivalent tissue (VTISS x λtiss) volumes.

$$VTOT = VLB + VTISS \times \lambda tiss$$

where VTISS = lung tissue volume

$\lambda tiss$ = solubility constant of N_2O in lung tissue.

4. The uptake of nitrous oxide by pulmonary blood ($\dot{V}N_2O$) was calculated as:

$$\dot{V}N_2O = VTCT_o \times FAN_2O_o - VTOT_{10} \times FAN_2O_{10}$$

where $VTCT_o$ = total rebreathing volume at time zero

$VTOT_{10}$ = " " " at time ten seconds

FAN_2O_o = mean alveolar concentration of N_2O at time zero

FAN_2O_{10} = mean alveolar concentration of N_2O at time ten seconds.

5. Cardiac output ($\dot{Q}c$) was calculated by the equation:

$$\dot{Q}c = \frac{\dot{V}N_2O}{\bar{FAN}_2O \times \lambda B}$$

where $\bar{FAN}_2O$ = the mean alveolar concentration of N_2O during the rebreathing period.

λB = solubility constant of N_2O in blood.

6. The mean alveolar concentration of N_2O ($\bar{FAN}_2O$) was calculated as the average mean alveolar end-expired concentration of N_2O at zero and ten seconds:

$$\bar{FAN}_2O = \frac{FAN_2O.o + FAN_2O.10}{2}$$

7. The expanded formula which was used to calculate cardiac output was therefore:

$$\dot{Q}c\ (l/min) = \frac{VTOT.o \times FAN_2O.o - VTOT.10 \times FAN_2O.10}{\frac{FAN_2O.o + FAN_2O.10}{2} \times \lambda B} \times 6$$

The term in the numerator is equivalent to the term $\dot{V}N_2O$ in equations 4 and 5, and is the uptake of N_2O by pulmonary blood, measured in litres. To measure the uptake in one minute, $\dot{V}N_2O$ is multiplied by 6.

The terms in the denominator are the mean alveolar N_2O concentration ($F\bar{A}N_2O$) during the rebreathing period (equations 5 and 6) and the solubility constant of N_2O in blood.

DISCUSSION OF THE APPARATUS AND TECHNIQUES

The nitrous oxide rebreathing method was modified to exclude possible sources of technical error and to make all measurements more rapid. The latter aim was largely achieved since all data was reproduced by the computer within half an hour. The cardiac output was calculated manually, however this was done out of choice to allow a more detailed analysis of the data. The procedure probably added a further 15 minutes for calculations which could have been performed on the off-line computer program.

The sources of technical error and their removal are discussed below.

SOURCES OF TECHNICAL ERROR: The use of the mass spectrometer and computer for breath by breath analysis of gas exchange has been examined by previous workers (Davies et al. 1974). This section examines the sources of error when the same equipment was used in the N_2O rebreathing procedure. Some of these errors were technical while others were errors in theoretical assumptions.

As discussed earlier, the measurement of an N_2O concentration on the mass spectrometer differed from the true reading by no more than 1% (e.g. 10% ± 1% of 10%). There was a further possible difference of 0.1% between the measured and the absolute readings of a specific gas tension (e.g. 5.0% ± 0.1%). However these were the largest possible errors in the mass spectrometer and were mostly eliminated by the computer, which also corrected small zero drift instabilities and changes in the pressure sensitivity of the spectrometer sampling tip. The result of these corrections was an extremely sensitive response to the measurement of nitrous oxide, so that in practice multiple readings of an N_2O concentration differed by no more than 0.01% N_2O (S.D.) of the correct reading.

In contrast the analysis of N_2O with the infra-red N_2O analyser (described in Section **b**) was liable to considerable error. Although the concentration of N_2O could be read to within 0.02% of a reading, it was achieved at great inconvenience to the user and was not the maximum error which could have been achieved with the normal use of the instrument. Despite the manufacturer's assurances, the analyser was sensitive to pressure gradients, and it was necessary to correct for these in the use and calibration of the instrument. Furthermore, although the instrument was stable it was not linear throughout its range, and its mechenical beam chopper also interfered with the recording of the N_2O concentration.

OTHER SOURCES OF ERROR 1) MEASUREMENT OF N_2O UPTAKE: During the rebreathing period the change in the N_2O concentration was not truly exponential, since there were simultaneous changes in lung-bag volume. Nevertheless, this did not alter the nature of the

semilogarithmic plot of N_2O; in the period between 4-10 seconds when N_2O concentrations were plotted, the slope of the curve was found to be linear in all studies (Figure 2.18). Changes in total rebreathing volume were recorded from the argon concentration throughout the rebreathing procedure. The reason for plotting the argon concentration on linear ordinates was discussed earlier. It was argued that the exponential loss of N_2O from the rebreathing system was balanced by an exponential release of CO_2 into the system. A residual removal of oxygen from the system occurred at a constant rate, and would be reflected in the linear slope of the argon concentration. Observations on 120 argon traces have borne out this assumption of linearity.

ERRORS IN MEASUREMENT OF MEAN ALVEOLAR N_2O CONCENTRATION: The mean alveolar concentration of nitrous oxide represented the driving pressure which forced N_2O into pulmonary blood. It was measured as the arithmetic average of the initial and final expired alveolar N_2O concentration in the period that N_2O uptake was calculated. The procedure had an intrinsic error since the mean alveolar N_2O ($F\bar{A}N_2O$) should represent this concentration for the whole rebreathing cycle (i.e. inspiration and expiration).

The true mean alveolar concentration of N_2O could be calculated from the inspiratory and expiratory alveolar concentrations of N_2O, plotted semi-logarithmically against time.

Using a record of an N_2O rebreathing procedure from the mass spectrometer (similar to Figure 2.10), the inspiratory alveolar N_2O

concentration could be obtained by extrapolating the slope of expired alveolar N_2O to the inspiratory phase of each breath (see the Ayotte method, Sectionb). The mean alveolar concentration of N_2O could then be taken as the average of the inspiratory and expiratory mean values of $F\bar{A}N_2O$, calculated from equation 6. Using a 10% N_2O mixture, the maximum difference between this $F\bar{A}N_2O$ and that calculated from expired breath was found to be 0.1%. In every rebreathing procedure this difference could be checked from the mass spectrometer and computer records (Table 2.3). The effect of the difference was a maximum underestimation of 0.3l/minute in cardiac output measured at high work loads. This error was considerably smaller than the error of planimetry.

It was noted earlier (Ayotte method, Ch. 2: Section b) that the technical error of measuring $F\bar{A}N_2O$ by planimetry produced a variation of $\pm$0.4l/min in cardiac output. However, the overall variation was greater than this due to a serious flaw in the theoretical assumption underlying the use of planimetry. In contrast the present method of measuring $F\bar{A}N_2O$ was rapid and the errors of all assumptions were readily analysed.

CHAPTER 2

(d) STATISTICAL NOTES

This thesis intended to investigate the reproducibility of several CO_2 and N_2O rebreathing methods for measuring cardiac output. In order to do this, these methods were examined during repeated use on individual subjects.

Both the experimental design, and the subsequent statistical analysis of the data were aimed at differentiating the sources of overall variation in the measurement of cardiac output. Two experimental procedures were generally adopted.

1. Repeated measurements of cardiac output were made on individual subjects during bicycle exercise at one or several work loads (Chapter 3, 5, 6 and 7).

2. Single daily measurements of cardiac output were repeated in individual subjects over a period of several days. On each day the subject exercised on a bicycle ergometer at one of several work loads (Chapter 4).

The reproducibility, or more correctly, the variation in estimating cardiac output was assessed by two statistical methods, both of which have in the past been called the "coefficient of variation".

METHOD 1. At each work load a mean value of cardiac output was calculated from the repeated measurements of cardiac output in each subject. The standard deviation about the mean was then assessed.

(1) The mean, $\bar{x} = \frac{\Sigma x}{n}$

where $\bar{x}$ = mean value of cardiac output for 1 subject at a given work load.

Σx = sum of the individual values of cardiac output at that work load.

n = number of observations.

(2) The standard deviation was then calculated:

$$SD = \sqrt{\frac{\Sigma x^2 - (\Sigma x)^2/n}{n - 1}}$$

where S.D. = standard deviation.

Σx^2 = sum of all individual squared values of cardiac output.

$\Sigma(x)^2$ = sum of all the values of cardiac output squared.

n = number of measurement of cardiac output on each subject.

(3) It was desirable to assess the variation in the estimate of cardiac output independently of absolute values of mean cardiac output. The coefficient of variation (C.V) or the standard deviation % of cardiac output was therefore calculated:

$$C.V. = \frac{S.D.}{\bar{x}} \times 100\%$$

where C.V. = coefficient of variation %, (or SD%)

S.D. = standard deviation of cardiac output

$\bar{x}$ = mean value of cardiac output

The coefficient of variation assessed the overall variation in the measurement of cardiac output. This was derived from both the technical errors in the method and the biological variation in subjects on whom the measurement was made.

METHOD 2: The reproducibility of a method of measuring cardiac output was also assessed from duplicate estimates in each of a group of subjects. The statistical result of the procedure has also been termed the coefficient of variation, or, the standard error of a single estimate. This method of analysis has been much used in the past to express the technical errors in a method (Holmgren and Pernow, 1950; Saltin, 1964; Becklake et al. 1962; Simmons and Shephard, 1971).

Paired estimates of cardiac output were taken from the repeated measurements in individual subjects. The duplicate estimates were selected so that either the penultimate or final pair of cardiac output values were taken from each of several work loads in individual subjects.

1. As before, the mean value of cardiac output ($\bar{x}$) was calculated for the individual values of cardiac output from all subjects at a given work load.

$$\bar{x} = \frac{\Sigma x}{n}$$

where $\bar{x}$ = mean value of cardiac output for all subjects at a given work load.

Σx = sum of all the values of cardiac output.

n = total number of observations.

2. The difference (d) between paired estimates of cardiac output in individual subjects was calculated, so that for all pairs x_1, x_2:

$$d = x_1 - x_2$$

3. The standard deviation of the difference (S.Dd) was calculated by the equation:

$$S.Dd = \sqrt{\frac{\Sigma d^2 - (\Sigma d)^2/n}{n - 1}}$$

where Σd^2 = sum of all squared differences.

$(\Sigma d)^2$ = sum of all differences squared.

n = number of observations.

Mathematically, it can be shown that the variance of the difference between paired estimates of cardiac output ($S.Dd^2$) is comprised of 1) the independent variance of the first and second value of cardiac output and 2) from a covariance or interaction term between the first and second estimates.

1) Variance of the difference = Variance of the 1st estimates of cardiac output + Variance of 2nd estimate of cardiac output + Covariance term

By making certain assumptions about the variance terms, the coefficient of variation of cardiac output could be estimated from the analysis of paired values. A first assumption was that the covariance term was most significant, since the two estimates of cardiac output were made independantly of each other. It was

further assumed that the independant variance in each of the paired values of cardiac output was the same.

2) Variance of the difference = Variance of the 1st estimate of cardiac output + Variance of the 2nd estimate of cardiac output

where

3) Variance of the difference = 2 x variance of a single estimate of cardiac output

The variance in a single estimate of cardiac output could therefore be assessed as:

4) $$\text{Variance of a single estimate of cardiac output} = \frac{\text{Variance of the difference}}{2}$$

For a single estimate of cardiac output the standard deviation measured from a number of paired observations is given by:

5) $$\text{S.D. (single estimate)} = \sqrt{\frac{\text{Variance of the difference}}{2}}$$

$$\text{or} = \sqrt{\frac{\text{S.Dd}}{2}}$$

6) The coefficient of variation of cardiac output (otherwise known as the standard error of a single estimate of cardiac output) was calculated from equation 5:

7) $$\text{Coefficient of Variation} = \frac{S.Dd}{\sqrt{2}\,\bar{x}} \times 100\%$$

To avoid confusion in the terminology and derivation of the above coefficient of variation, it will be called the coefficient of variation from a paired estimate (C.V.P.E.) throughout this thesis.

The C.V.P.E. index has been used by most workers to assess the technical error in the method of measuring cardiac output.

By measuring the difference in paired estimates of cardiac output in a single subject, it is assumed that the biological variation inherent in both the estimates is eliminated to a large degree. However, this is obviously dependant on the time interval between duplicate estimates.

Elimination of the biological variation within subjects leaves the residual technical variation of the method. The difference in the variation between subjects is not considered in the calculations, however, it does affect the estimate of the C.V.P.E. since it alters the mean value of cardiac output in the group (the denominator in equation 7).

In this thesis both the coefficient of variation (C.V.) and the coefficient of variation from paired estimates (C.V.P.E.) were used to analyse the reproducibility of several methods of measuring cardiac output. The two coefficients are in no way related mathematically;

however they are both used here to allow a direct comparison with previous studies in the literature.

The use of the C.V. and C.V.P.E. indices in the measurement of overall and technical variation of a method is discussed in the relevant sections of the thesis. Their use has allowed a comparison of the reliability of the two methods.

C.V.P.E.: The success of the C.V.P.E. depends on the degree to which the biological variation within and between subjects is eliminated by taking the difference of paired estimates of cardiac output. The elimination of biological variation within subjects depends on the time interval between the duplicate estimates. It also depends on the range of biological variation in a group of subjects in a given time interval. If the technical variation of the method is assumed constant, then this range is indicated in the coefficients of variation (C.V.) of cardiac output for each subject (Chapters 4, 5, 6 and 7). The C.V. shows that each individual has a particular level of biological variation which affects the C.V.P.E. index. Finally, the C.V.P.E. is also affected by differences in the absolute value of cardiac output between subjects; since this affects the mean value ($\bar{x}$) of cardiac output, the value of C.V.P.E. will therefore depend on the homogeneity of the studied group.

C.V.: The coefficient of variation (C.V.) provides an index of overall variation for individual subjects and depends on multiple (as opposed to paired) estimates of cardiac output on each subject. Whereas the C.V.P.E. gives a single index of variation for a group of

subjects, the C.V. gives a range. The range of C.V. in a group of individuals is obviously affected by the mean value of cardiac output in each subject (S.D./mean), but this can be assessed statistically provided the relative contribution of each mean is assessed by using the "weighted" mean. This makes the C.V. a statistically more reliable index of variation since it does not depend on broad assumptions of the C.V.P.E. Nevertheless, in the present work the two variation coefficients are used as complementary indices of variation.

COMPARISON OF CARDIAC OUTPUT MEASURED BY DIFFERENT METHODS:

In the present work non-invasive measurements of cardiac output were generally made on a group of subjects who exercised at three or more levels of work intensity. It was often desirable to compare the absolute values of cardiac output at each level of work with the values found by other methods.

For a given method the estimates of cardiac output were treated to simple linear regression analysis, in which oxygen consumption was the independant variable. Standard procedures were used for the analysis, as described by Armitage (1971).

The use of a linear regression to the measurements of cardiac output is however open to some doubt. Many workers have found that during exercise, cardiac output increases linearly with oxygen consumption. Nevertheless, at the two extremes of rest and very high work loads the linearity is not certain. At rest the variability of any method of measuring cardiac output is too high to allow any

firm conclusions to be drawn from regression analysis. At very high work loads the slope of cardiac output against oxygen consumption tends to flatten.

In view of these considerations the linear regression analysis of cardiac output was restricted to measurements made during submaximal exercise.

CHAPTER 3

A COMPARISON OF LINEAR AND EXPONENTIALLY EXTRAPOLATED VALUES OF MIXED VENOUS PCO_2

INTRODUCTION

The plateau CO_2 rebreathing method of Collier (1956) and the exponential CO_2 rebreathing method of Defares (1958) are both used to measure mixed venous PCO_2.

In a recent study of normal subjects Godfrey and Wolf (1972) compared the plateau and exponential rebreathing methods during exercise, and found that the plateau method gave more reproducible estimates of $P\bar{v}CO_2$. Their analysis of the literature also indicated that the exponential method as used by various workers (Jernerus et al. 1963: Ferguson et al. 1968) was not very reproducible.

However, there are several graphical exponential solutions for mixed venous PCO_2 (Jernerus et al. 1963; Klausen, 1965; Defares, 1958; 1972), while in the plateau method, a linear extrapolation procedure can be used to estimate $P\bar{v}CO_2$ in the absence of a plateau $P\bar{v}CO_2$ (Denison, 1968).

The present study set out to investigate the use of these extrapolation procedures in the estimation of mixed venous PCO_2.

SUBJECTS and METHODS

Two similar studies were performed on groups of six and seven healthy volunteers. Anthropometric data are presented in Table 3.1. The first three subjects (EZF, MR, PF) took part in both studies.

Table 3.1: Anthropometric data of the two groups of normal subjects in the study.

SUBJECT	AGE (yrs)	SEX	Ht (cm)	Wt (kg)
		GROUP 1		
EZF	23	M	173	70.0
MR	30	M	180	75.5
PF	33	M	178	73.5
SS	23	M	183	80.6
JP	28	M	187	73.2
JZ	16	M	169	59.2
HP	17	M	165	57.5
		GROUP 2		
EZF	23	M	173	70.0
MR	30	M	180	75.5
PF	33	M	178	73.5
PK	34	M	164	62.5
ML	27	M	174	70.0
AA	22	M	170	61.0

Each subject sat on a bicycle ergometer and exercised at a constant power output of 75 watts for a period of 25-30 minutes. Heart rate was recorded electrocardiographically and steady state measurements of ventilation and gas exchange were made at intervals of five minutes, as outlined in Chapter 2 (Section: a).

At five minute intervals the subject rebreathed from a bag containing a known concentration of carbon dioxide in oxygen. Alveolar PCO_2 was monitored continuously at the mouth during this period. At the end of each rebreathe, steady state measurements of heart rate, ventilation and gas exchange were continued.

In the first study, five rebreathing manoeuvres were carried out with five gas mixtures containing 0, 3, 6, 8 and 14% CO_2 in oxygen, given in random order. The 14% CO_2 mixture gave a true mixed venous PCO_2 plateau in all the subjects at this work load. In the second study, mixtures of 8, 10, 12, 14, 16 and 18% CO_2 in oxygen were used.

Mixed venous PCO_2 was calculated in a variety of ways, based either on a plateau rebreathing technique using the linear extrapolation procedure (Denison, 1968), or on the exponential rebreathing techniques as used by Defares (1958, 1972), Klausen (1965) and Jernerus et al. (1963). These methods have been discussed fully in Chapter 2 (The CO_2 rebreathing method).

RESULTS

The results from the two studies are given in the Appendix, Table A3.1.

For each subject the oxygen consumption was measured during steady state conditions, before a rebreathing procedure was carried out with one of a number of CO_2 gas mixtures (shown in Table A3.1). For each rebreathe several estimates of $\bar{P}vCO_2$ were calculated by the linear or exponential graphical solutions, as outlined earlier.

The reproducibility of five different exponential solutions for $\bar{P}vCO_2$ was assessed from the data in Table A 3.1. It was found (Table 3.2) that the method of Klausen (1965) was the most reproducible, with a mean coefficient of variation of 15.4% for the group. The method of Jernerus et al. (1963) was the least reproducible. The method of Klausen was therefore selected for comparison against the linear extrapolation method of Denison. The values of $\bar{P}vCO_2$ based on the linear (Denison) and exponential (Klausen) extrapolations were used to calculate the cardiac output in each subject. Some of the exponential results for $\bar{P}vCO_2$ (even by the Klausen method) were either below the calculated $PaCO_2$, or were in excess of 100 mmHg. These values would have given negative or unrealistically low values of cardiac output and were therefore omitted from Table A 3.1.

In each subject $\bar{P}vCO_{2s}$ measured by the linear (Denison) and exponential (Klausen) extrapolation procedures are shown in Tables 3.3 and 3.4 respectively. Each table is divided in two halves which represent two separate studies.

Table 3.2: Showing the mean coefficient of variation of $\bar{P}v\bar{C}O_2$ ($\pm$ 1 S.D.) for the group, estimated from a range of CO_2 rebreathing mixtures.

	CO_2 REBREATHING METHOD OF:				
	KLAUSEN	KLAUSEN/ DEFARES	DEFARES	JERNERUS/ DEFARES	JERNERUS
Mean coefficient of variation of $\bar{P}v\bar{C}O_2$ for the group (C.V.%)	15.4	15.5	16.9	20.5	40.1
S.D. of the mean coefficient of variation	$\pm$ 12.7	$\pm$ 11.8	$\pm$ 15.1	$\pm$ 20.8	$\pm$ 26.0

Table 3.3: Presents the values of $P\bar{v}CO_2$ measured by the Denison (linear) extrapolation procedure in each subject. The mean values ($\pm$ 1 S.D.), and the coefficients of variation of $P\bar{v}CO_2$, cardiac output and oxygen consumption are also presented for each subject, and for the group.

T A B L E 3.3: S T U D Y - 1

SUBJECT	DENISON (LINEAR) EXTRAPOLATION of $P\bar{v}CO_2$ (mmHg) REBREATHING CO_2 CONCENTRATION					FOR INDIVIDUAL SUBJECTS								
						MIXED VENOUS PCO_2 (mmHg)			CARDIAC OUTPUT (l/min)			OXYGEN CONSUMPTION (ml/min)		
	0%	3%	6%	8%	P	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%
EZF	64	61	62	59	60	61	2	3.1	10.7	0.5	4.2	1299	81	6.2
MR	57	56	60	61	64	60	3	5.4	11.8	1.9	16.1	1309	51	3.9
PF	64	64	66	66	69	66	2	3.0	10.8	0.4	4.3	1271	25	1.9
SS	61	64	66	67	66	65	2	3.3	9.8	0.9	9.5	1184	100	8.4
JP	66	67	63	61	65	64	2	3.9	12.9	0.5	3.9	1270	27	2.1
JZ	61	64	63	64	66	64	2	2.7	11.2	1.0	8.9	1266	49	3.8
HP	63	63	63	65	68	64	2	3.7	10.9	0.9	8.3	1231	36	2.9
	FOR the GROUP													
MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	62	63	63	63	65									
MIXED VENOUS PCO_2 (mmHg) SD	3	3	2	3	3									
MIXED VENOUS PCO_2 (mmHg) CV%	4.7	5.4	3.1	4.6	4.6									
CARDIAC OUTPUT (l/min) $\bar{m}$	11.4	11.9	10.9	10.9	10.1									
CARDIAC OUTPUT (l/min) SD	1.5	1.3	1.5	1.1	1.0									
CARDIAC OUTPUT (l/min) CV%	12.8	10.9	13.5	10.1	9.8									
OXYGEN CONSUMPTION (ml/min) $\bar{m}$	1278	1281	1242	1249	1257									
OXYGEN CONSUMPTION (ml/min) SD	104	64	77	17	52									
OXYGEN CONSUMPTION (ml/min) CV%	8.1	5.0	6.1	1.3	4.1									

TABLE 3.3 (continued): STUDY - 2

SUBJECT	DENISON (LINEAR) EXTRAPOLATION of $P\bar{v}CO_2$ (mmHg) REBREATHING CO_2 CONCENTRATION						FOR INDIVIDUAL SUBJECTS: MIXED VENOUS PCO_2 (mmHg)			CARDIAC OUTPUT (l/min)			OXYGEN CONSUMPTION (ml/min)		
	8%	10%	12%	P	16%	18%	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%
EZF	64	65	64	64	64	66	64	1	1.2	11.1	0.5	4.0	1385	86	6.2
MR	63	62	64	65	66	68	65	2	3.7	13.1	0.9	6.9	1322	31	2.3
PF	66	67	67	72	73	75	70	4	5.2	10.1	1.1	11.2	1292	23	1.7
PK	58	58	64	62	63	62	61	3	4.4	10.3	0.6	4.9	1176	34	2.8
ML	58	59	61	64	63	63	61	2	3.6	12.0	0.8	6.3	1181	27	2.3
AA	64	62	64	63	—	63	63	1	0.9	13.0	1.1	8.2	1319	88	6.6

FOR the GROUP

MIXED VENOUS PCO_2 (mmHg)	$\bar{m}$	62	62	64	65	66	66
	SD	3	3	2	4	4	5
	CV%	5.2	5.4	3.4	5.7	6.1	7.3
CARDIAC OUTPUT (l/min)	$\bar{m}$	12.2	12.2	11.5	11.3	10.9	11.2
	SD	1.7	1.7	1.0	1.3	1.5	1.5
	CV%	14.3	13.6	8.2	12.1	13.6	13.4
OXYGEN CONSUMPTION (ml/min)	$\bar{m}$	1278	1282	1250	1283	1281	1299
	SD	99	75	109	89	99	117
	CV%	7.4	5.8	8.7	6.9	7.7	9.0

Table 3.4: Values of $P\bar{v}CO_2$ measured by the Klausen (exponential) extrapolation procedure in each subject. The mean values ± 1 S.D.), and the coefficients of variation of $P\bar{v}CO_2$, cardiac output and oxygen consumption are also shown for each subject, and for the group.

T A B L E 3.4: S T U D Y - 1

SUBJECT	KLAUSEN (EXPONENTIAL) EXTRAPOLATION of $P\bar{v}CO_2$ (mmHg) REBREATHING CO_2 CONCENTRATION 0%	3%	6%	8%	P	FOR INDIVIDUAL SUBJECTS MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	SD	CV%	CARDIAC OUTPUT (l/min) $\bar{m}$	SD	CV%	OXYGEN CONSUMPTION (ml/min) $\bar{m}$	SD	CV%
EZ	63	69	74	58		66	7	10.3	9.8	2.1	21.3	1299	81	6.2
MR	—	98	68	79		82	15	18.1	7.2	1.4	20.0	1309	51	3.9
PF	86	99	92	66		86	14	16.7	6.7	2.2	32.0	1271	25	1.9
SS	64	67	—	73		82	27	33.0	8.9	1.0	10.6	1184	100	8.4
JP	74	66	85	—		75	9	12.4	10.2	3.5	34.6	1270	27	2.1
JZ	55	60	63	78		64	10	15.8	12.4	3.6	29.3	1266	49	3.8
HP	56	70	66	65		64	6	9.1	11.1	2.2	20.1	1231	36	2.9
	FOR the GROUP													
MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	66	76	75	70										
MIXED VENOUS PCO_2 (mmHg) SD	12	16	12	8										
MIXED VENOUS PCO_2 (mmHg) CV%	17.7	21.2	15.4	11.7										
CARDIAC OUTPUT (l/min) $\bar{m}$	10.9	9.6	8.6	9.2										
CARDIAC OUTPUT (l/min) SD	3.5	3.7	2.3	2.0										
CARDIAC OUTPUT (l/min) CV%	32.3	38.4	26.5	21.3										
OXYGEN CONSUMPTION (ml/min) $\bar{m}$	1278	1281	1242	1249										
OXYGEN CONSUMPTION (ml/min) SD	104	64	77	17										
OXYGEN CONSUMPTION (ml/min) CV%	8.1	5.0	6.1	1.3										

TABLE 3.4 (continued): STUDY - 2

SUBJECT	KLAUSEN (EXPONENTIAL) EXTRAPOLATION of $\bar{P}vCO_2$ (mmHg) REBREATHING CO_2 CONCENTRATION						FOR INDIVIDUAL SUBJECTS								
							MIXED VENOUS PCO_2 (mmHg)			CARDIAC OUTPUT (l/min)			OXYGEN CONSUMPTION (ml/min)		
	8%	10%	12%	P	16%	18%	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%	$\bar{m}$	SD	CV%
EZF	66	70	—		65	72	68	3	4.9	9.8	0.5	4.6	1385	86	6.2
MR	62	69	63		70	71	67	4	6.1	12.0	2.4	20.2	1322	31	2.3
PF	69	80	72		76	80	76	5	6.3	8.8	1.3	15.1	1292	23	1.7
PK	56	84	62		65	67	67	11	15.7	9.3	1.8	18.9	1176	34	2.8
ML	—	65	58		66	71	65	5	7.9	10.9	1.5	13.7	1181	27	2.3
AA	61	53	64		—	74	63	8	13.0	11.9	2.0	16.8	1319	88	6.6
	FOR the GROUP														
MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	63	70	64		68	72									
MIXED VENOUS PCO_2 (mmHg) SD	5	11	5		4	4									
MIXED VENOUS PCO_2 (mmHg) CV%	8.0	15.8	8.0		6.4	6.0									
CARDIAC OUTPUT (l/min) $\bar{m}$	12.2	9.1	11.6		9.7	9.3									
CARDIAC OUTPUT (l/min) SD	2.5	2.1	1.3		0.9	1.1									
CARDIAC OUTPUT (l/min) CV%	20.9	23.3	11.2		8.9	12.1									
OXYGEN CONSUMPTION (ml/min) $\bar{m}$	1278	1282	1250		1281	1299									
OXYGEN CONSUMPTION (ml/min) SD	99	75	109		99	117									
OXYGEN CONSUMPTION (ml/min) CV%	7.7	5.8	8.7		7.7	9.0									

The mean $\bar{P}vCO_2$ ($\pm$1 S.D.) and its coefficient of variation is given for each subject rebreathing a range of gas mixtures. Similar data is given for the oxygen consumption and cardiac output (based on individual estimates of $\bar{P}vCO_2$).

For all subjects rebreathing the same CO_2 gas mixture, Tables 3.3 and 3.4 present the mean values ($\pm$1 S.D.) of $\bar{P}vCO_2$, cardiac output and oxygen consumption, together with their coefficients of variation.

The mean values of $\bar{P}vCO_2$ ($\pm$1 S.D.) for the group were plotted against both the initial CO_2 content of the rebreathing bag and the mean oxygen consumption (Figure 3.1). Linearly extrapolated estimates of $\bar{P}vCO_2$ were relatively reproducible throughout the range of rebreathing mixtures used. In comparison, the values of $\bar{P}vCO_2$ based on the exponential procedure of Klausen became more reproducible only when the CO_2 content of the rebreathing bag was raised towards the plateau equilibrium mixture (14% CO_2 in oxygen).

The variations in $\bar{P}vCO_2$ were reflected in the group mean values of cardiac output, which are shown in Figure 3.2. The values of cardiac output based on linearly extrapolated $\bar{P}vCO_2$ were consistent throughout the range of rebreathing mixtures (standard deviation of $\dot{Q}$ = $\pm$ 1.5 l/min.). In comparison, the group mean values of cardiac output based on exponentially extrapolated $\bar{P}vCO_2$ varied by as much as $\pm$3.5 l/min (S.D.) when low CO_2 mixtures were used. The cardiac output values became more consistent only when the CO_2 concentrations in the rebreathing mixture was raised.

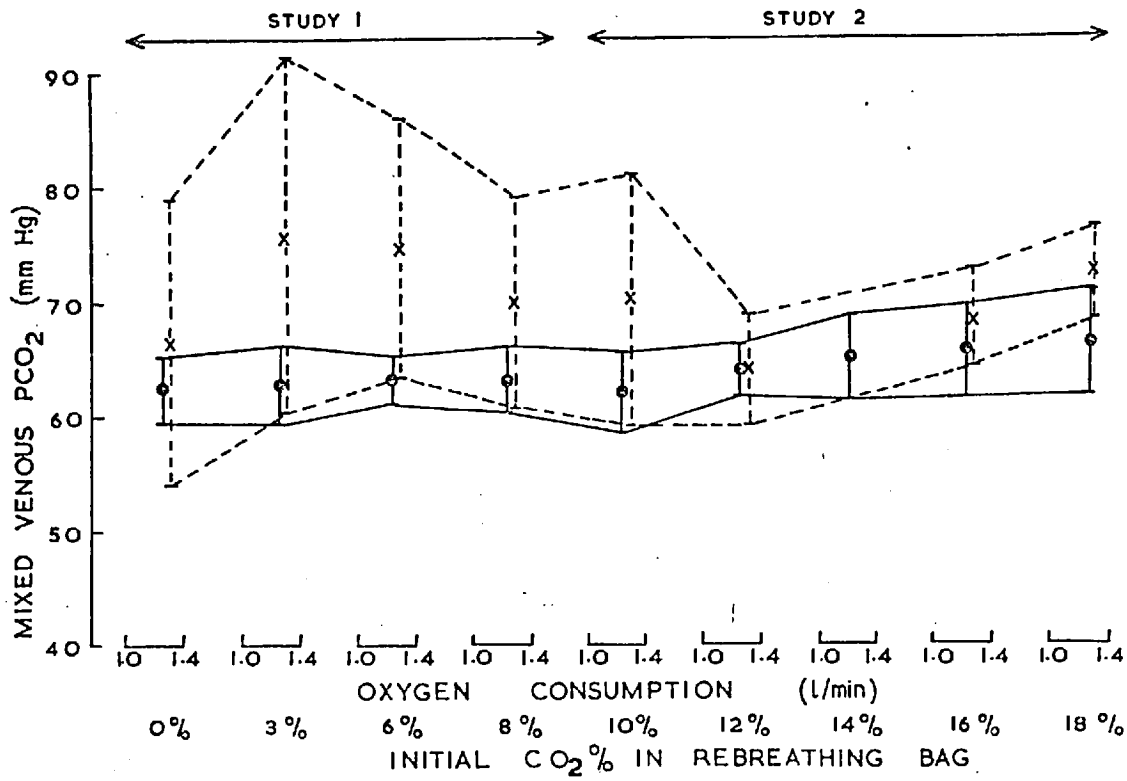


Figure 3.1 Mean values of $P\bar{v}CO_2$ (± 1S.D) in the group, determined by the linear and exponential extrapolation procedures, are plotted against (i) the mean oxygen consumption of the group prior to the rebreathing manouvre, and (ii) the initial CO_2 content of the rebreathing mixture used to determine $P\bar{v}CO_2$.

(Plateau & Linearly extrapolated values: ——o——)
(Exponentially extrapolated values: ----x----)

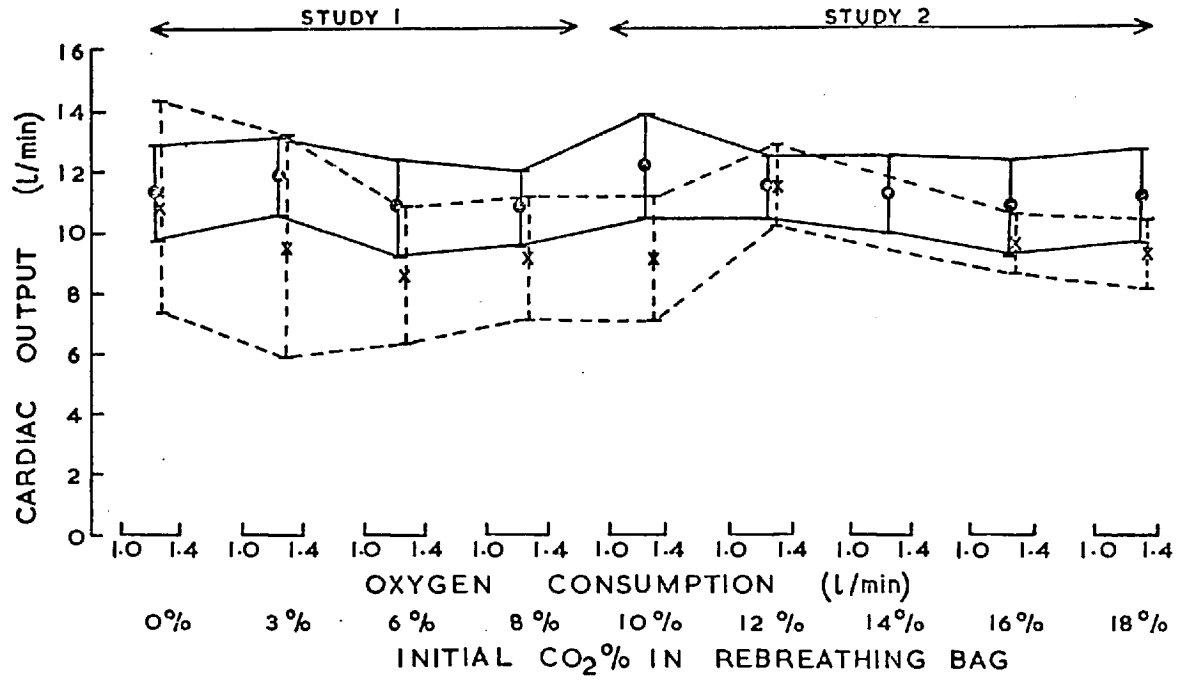


Figure 3.2 Mean values of cardiac output in the group (±1S.D) based on linear and exponential extrapolates of $P\bar{v}CO_2$, plotted against (i) the mean oxygen consumption in the group, and (ii) the initial CO_2 content of the rebreathing mixture.
(Plateau & Linearly extrapolated values : ——o——)
(Exponentially extrapolated values: ----x----)

DISCUSSION

In both the linear and exponential rebreathing methods the mean values of cardiac output in the group (Figure 3.2) were within the 95% confidence limits for cardiac output in normal subjects, as measured by the direct (O_2) Fick method (Holmgren and Pernow, 1960: Bevegard et al. 1960). However, there are marked differences in the standard deviations of cardiac output based on the two rebreathing methods, and these obviously reflect the variation in the estimates of mixed venous PCO_2.

THE EXPONENTIAL EXTRAPOLATION OF $P\bar{v}CO_2$: It was noted earlier that various exponential graphical solutions were used to estimate $P\bar{v}CO_2$ (Table 3.2). The method of Kalusen and its modification (Klausen/Defares method) gave the most reproducible estimates of $P\bar{v}CO_2$. This method differed from those of Defares and Jernerus principally in the use of an initial tension-time curve for the alveolar PCO_2 during the rebreathing period. It therefore seems that the use of a tension-time curve is desirable for the reproducibility of results. The tension-time curve merely standardises the exponential rise of alveolar PCO_2 with respect to time. It is notable that the Defares method (1972), which plots the rate of change of alveolar PCO_2 against the alveolar PCO_2 of the initial breath, (thus standardising the time interval between breaths) achieves a reproducibility which is intermediate between the Klausen and Jernerus methods. The method of Jernerus (and the modified Jernerus/Defares method) use no standardisation technique for time, and are also the least reproducible. According to Defares (1972) the use

of a tension-time curve is undesirable, as it causes a subsequent under-estimation of $P\bar{v}CO_2$. However, Defares does not give evidence to support this statement, so it is difficult to comment on his observation. Had Defares been correct then the present measurements would have under-estimated $P\bar{v}CO_2$. However, the mean values of $P\bar{v}CO_2$ in the Klausen method were high when compared with both linearly extrapolated (and plateau) estimates of $P\bar{v}CO_2$ in this study (Figure 3.1), and with values in the literature (Jones et al. 1967; Higgs et al. 1967; Denison, 1968).

Although the rebreathing mixtures in this study spanned a range of 0-18% CO_2 in oxygen, a 3-6% CO_2 in oxygen mixture is generally used to measure $P\bar{v}CO_2$ by the exponential methods (Defares, 1958, 1972; Jernerus et al. 1963; Klausen, 1965; Ferguson et al. 1968; Muiesan et al. 1968). Within this range, the authors who have proposed the various graphical solutions for $P\bar{v}CO_2$ have often quoted reproducibility data which is insufficient, and which makes a comparison between methods difficult.

There is no readily available data on the reproducibility of the Defares rebreathing methods (1958, 1972). Similarly, Jernerus et al. (1963) do not quote the reproducibility of $P\bar{v}CO_2$ measured in individual subjects or in a group. However, where the mean value of cardiac output in each of six subjects is given, this varies from the group value by as much as $\pm$ 2.2 to 4.2 l/min (S.D.) during exercise. In a comparison of the exponential and plateau rebreathing methods, Godfrey and Wolf (1972) investigated the reproducibility of the Jernerus method and found a coefficient of

variation of 4.7 - 6.6% for $P\bar{v}CO_2$ measured in individual subjects during exercise. This is slightly higher than the 1.4 - 4.9% coefficient of variation found in a single subject using the Klausen method during exercise (Klausen, 1963). This agrees with the findings of this study, which also suggest that the Klausen method is more reproducible than the method of Jernerus. The present study also indicates that when the Klausen method is used once only in a group of subjects (using a 3-6% CO_2 in O_2 mixture) the coefficient of variation of $P\bar{v}CO_2$ is between 15-21% (Table 3.4).

THE PLATEAU AND LINEARLY EXTRAPOLATED $P\bar{v}CO_2$: Only one linear extrapolation procedure has been proposed for the estimation of $P\bar{v}CO_2$, and was originally used by Denison (1968) to supplement the plateau rebreathing procedure only when a plateau $P\bar{v}CO_2$ was unobtainable. Most of the literature has therefore concentrated on the use of the plateau technique, with little emphasis on the extrapolation procedure. Denison (1968) found that the extrapolated $P\bar{v}CO_2$ differed by no more than $\pm$ 1 mmHg from the plateau value over a wide range of initial rebreathing mixtures.

For the plateau method, a mean coefficient of variation of 0.8 - 1.4% has been found for $P\bar{v}CO_2$ measured in individual subjects during a period of exercise (Godfrey and Wolf, 1972). In the subjects of this study, the mean coefficient of variation of $P\bar{v}CO_2$ extrapolated from a range of low (0-8% CO_2 in O_2) or high (8-16% CO_2 in O_2) rebreathing mixtures was only 3.2 - 3.6%. The 2% difference in excess of the 0.8 - 1.4% variation from the data of Godfrey and Wolf (1972) is probably due to the use of the extrapolation procedure.

Higgs et al. (1967) found that the coefficient of variation of plateau $\bar{P}vCO_2$ measured once only in a group of exercising subjects was about 3-8%. Compared with this, the coefficient of variation of extrapolated $\bar{P}vCO_2$ in the present group was 3-7% (Table 3.3), indicating no marked loss of reproducibility when the extrapolation procedure was used once only in a group of subjects.

COMPARISON OF LINEAR AND EXPONENTIALLY EXTRAPOLATED ESTIMATES OF $\bar{P}vCO_2$:

In addition to the standard CO_2 in O_2 mixtures which are used for the plateau and exponential rebreathing procedures, a much wider range was used in this study, in order to test the flexibility of these methods.

The linear extrapolation gave consistently reproducible values of $\bar{P}vCO_2$ throughout the range of rebreathed gas mixtures. The coefficient of variation of $\bar{P}vCO_2$ in the group was always between 3-7% (Table 3.3) and its magnitude was unrelated to the initial concentration of CO_2 in the bag. This observation is particularly important in the context of a study by Ashton and McHardy (1963), which pre-dated the later criteria for the definition of a plateau $\bar{P}vCO_2$ (Jones et al. 1967; Denison, 1968). These workers found that their "plateau" $\bar{P}vCO_2$ was raised as the CO_2 content of the rebreathing mixture was increased. However, subsequent work (Jones et al. 1967; Denison, 1968) redefined a plateau as existing between 6-10 seconds during a rebreathing period and introduced the linear extrapolation procedure (Denison, 1968). The consistently low coefficients of variation of $\bar{P}vCO_2$ in the group (Table 3.3) suggest

that the extrapolations and plateau methods are both successful in eliminating the dependance of $\bar{P}vCO_2$ on the initial CO_2 content of the rebreathing mixture.

In contrast, alteration of the CO_2 content of the rebreathing bag had a marked effect on the reproducibility of $\bar{P}vCO_2$ estimated by the Klausen exponential extrapolation (also true of all the other exponential extrapolations). When a 100% oxygen mixture was rebreathed the coefficient of variation of $\bar{P}vCO_2$ in the group was 18% (Table 3.4). The group variation (C.V.) was reduced to 6% when a 16-18% CO_2 in oxygen mixture was rebreathed. The variation was therefore reduced as the CO_2 content of the rebreathing mixture was raised towards the plateau equilibration value (14% CO_2 in oxygen).

The observations of this study suggest several procedures for the measurement of $\bar{P}vCO_2$. Mixed venous PCO_2 should preferentially be measured by the plateau rebreathing method. In non-plateau conditions the linear extrapolation procedure could be used with more accuracy (and convenience) than the exponential extrapolation procedure. Although the use of an extrapolation (in place of a plateau) appears to affect the reproducibility of the $\bar{P}vCO_2$ estimate slightly, in single measurements of $\bar{P}vCO_2$ in a group of subjects the observed 3-7% variation is very similar to the 3-8% variation in plateau $\bar{P}vCO_2$ measured in a group (Higgs et al. 1967).

If the exponential extrapolation is to be used, two problems become evident. The recommended rebreathing mixture of 3-6% CO_2

in oxygen causes a considerable variation in the estimate of $P\bar{v}CO_2$, and could probably be reduced by altering the mixture to suit a particular work load. However, raising the initial CO_2 concentration achieves a reproducibility which is no better than that of the plateau rebreathing procedure.

CONCLUSIONS

The linear extrapolation procedure for estimating $P\bar{v}CO_2$ was consistently reproducible throughout a range of CO_2 rebreathing mixtures. In contrast the exponential rebreathing procedure was less reproducible and was associated with a number of problems. A comparison of several graphical solutions indicated that it was necessary to standardise the exponential rise of alveolar PCO_2 during the rebreathe with respect to time. The estimates of $P\bar{v}CO_2$ were also considerably affected by the initial CO_2 content of the rebreathing mixture.

It is therefore concluded that the plateau rebreathing procedure should be used as the method of choice. In the absence of a true plateau, a linear extrapolation should be used to estimate $P\bar{v}CO_2$. If the exponential rebreathing procedure is used, the initial rebreathing mixture should be altered in relation to the expected value of $P\bar{v}CO_2$ in the subject.

CHAPTER 4

THE REPRODUCIBILITY OF MIXED VENOUS PCO_2 AND CARDIAC OUTPUT MEASURED BY THE INDIRECT FICK CO_2 REBREATHING METHOD

INTRODUCTION

In the previous chapter it was found that the mixed venous PCO_2 measured by the plateau CO_2 rebreathing method was more reproducible than the value estimated by the exponential rebreathing method.

Several studies have examined the reproducibility of the plateau rebreathing method, however these have generally noted the variation in repeated measurements of $P\bar{v}CO_2$ made on a single occasion (Denison, 1968; Godfrey and Wolf, 1972), while none have assessed the day to day variation of the measurement.

The present study was therefore carried out to investigate the reproducibility of the plateau rebreathing method in subjects who were exercised repeatedly over a period of several days.

SUBJECTS and METHODS

The studies were performed by ten normal subjects, comprised of six men, one woman and three children. Some anthropometric data are presented in Table 4.1.

The subjects were seated on a bicycle ergometer and were initially at rest. They were then exercised at two or three work loads which were between $\frac{1}{4}$ and $\frac{3}{4}$ of each subject's previously determined maximum

Table 4.1: Anthropometric data of the normal subjects examined in the study.

SUBJECT	AGE (yrs)	SEX	Ht (cm)	Wt (kg)
1	22	M	173	66.0
2	27	M	171	69.0
3	27	M	176	81.5
4	19	M	177	67.0
5	30	M	167	67.0
6	17	M	182	63.5
7	26	F	165	52.5
8	15	M	156	43.5
9	14	M	154	43.3
10	12	M	151	34.0

working capacity (Wmax). The studies were carried out at one to three day intervals, and each subject completed at least four studies which included all work loads. The work loads were performed consecutively without pause, as outlined in Chapter 2 (CO_2 rebreathing method). Of ten subjects, three completed more than ten full studies. Two completed six, three others completed five, and the remaining two completed four studies each.

During each test, the subject's heart rate, ventilation and gas exchange were monitored continuously. Once the subject had reached a steady state in these parameters, oxygenated mixed venous PCO_2 was estimated by the plateau CO_2 rebreathing procedure. A true plateau $\bar{P}vCO_2$ was obtained in most of the rebreathes, and the linear extrapolation procedure of Denison (1968) was only needed in a few cases (32 out of 271 estimates of $\bar{P}vCO_2$). At the end of the rebreathing period the work load was raised and all measurements were repeated at the higher work load.

CALCULATIONS: The results were calculated as described earlier (Chapter 2), using a computer program devised by Godfrey (1970).

RESULTS

A statistical summary of all the measurements which were made in individual subjects is given in Table A-4.1 of the appendix. At each work load the mean values of various parameters are given, together with their coefficients of variation (C.V.) for each subject.

The oxygen consumption for a given work load was similar in all subjects (Figure 4.1), as was the ventilation at a given oxygen

consumption (Figure 4.2). Despite differences in the mixed venous PCO_2 between subjects (Figure 4.3), the cardiac output for a given oxygen consumption was also similar in all subjects (Figure 4.4). The heart rate (Figure 4.5) and stroke volume (Figure 4.6) were size and sex dependant, so that at a given oxygen consumption, subjects 6-10 (female and children) had higher heart rates and smaller stroke volumes than the adult male subjects (1-5). The within subject coefficients of variation of all parameters (C.V.) were high at rest, but fell during exercise (Figures 4.7, 4.8, 4.9 4.10, 4.11, 4.12). At a given oxygen consumption the coefficients of variation of heart rate in subjects 6-10 (women and children) were lower than those of subjects 1-5 (Figure 4.11). Since the standard deviations of the mean heart rate in subjects 6-10 were only slightly lower than those of the adults, the lower coefficients of variation were in effect due to the higher heart rates of these subjects at a given oxygen consumption (since C.V. = standard deviation x 100/mean). The difference in the coefficients of variation of heart rate was eliminated when these were plotted against the absolute mean heart rate in each subject (Figure 4.13).

The coefficient of variation of paired estimates of cardiac output (C.V.P.E.) is shown in Table 4.2. The index is calculated for all subjects working at similar oxygen consumptions, and attempts to assess the technical variation of the CO_2 rebreathing method (discussed later). The C.V.P.E. is considerably smaller than the overall coefficient of variation (C.V.) of cardiac output which is shown in Figure 4.10.

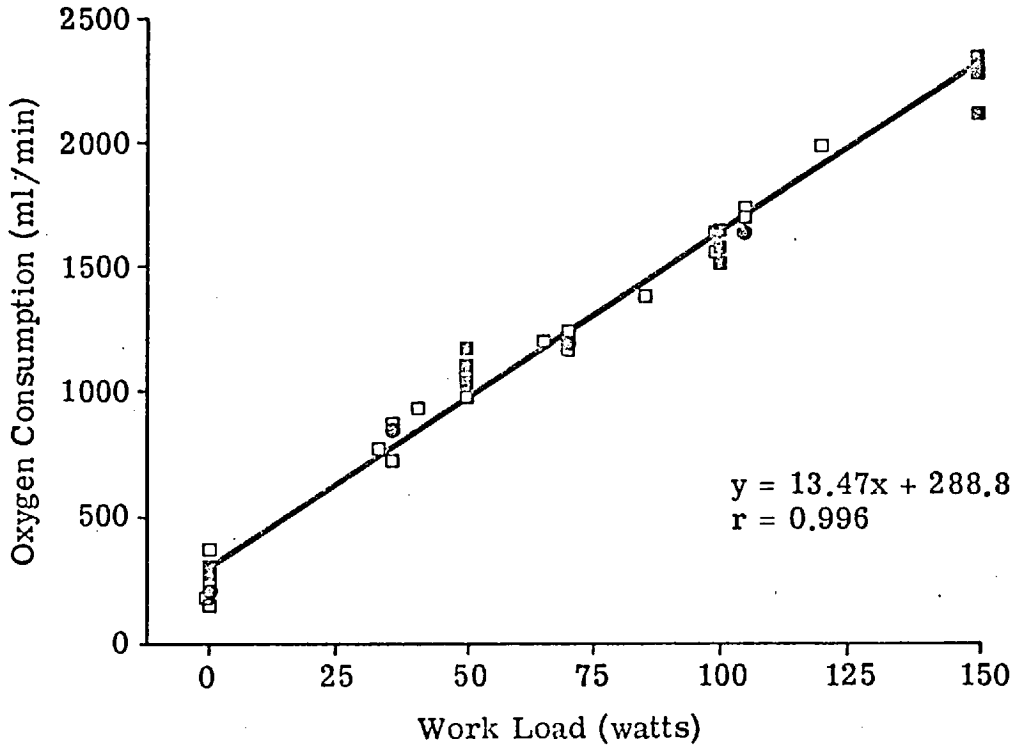


Figure 4.1 Shows the regression line for the mean oxygen consumptions of ten normal subjects, plotted against fixed work loads (usually 0, $\frac{1}{4}$, $\frac{1}{2}$, $\frac{3}{4}$ Wmax) performed on a bicycle ergometer in the sitting position.

Figures 4.2-4.6 The mean values of ventilation (4.2), plateau $P\bar{v}CO_2$ (4.3), cardiac output (4.4), heart rate (4.5), and stroke volume (4.6) in ten subjects are plotted against the corresponding mean oxygen consumptions at rest and during exercise. Regression lines for the group data are presented for ventilation and cardiac output (figures 4.2 and 4.4).

Adult males ◘ ; Adult female ● ; Children □ .

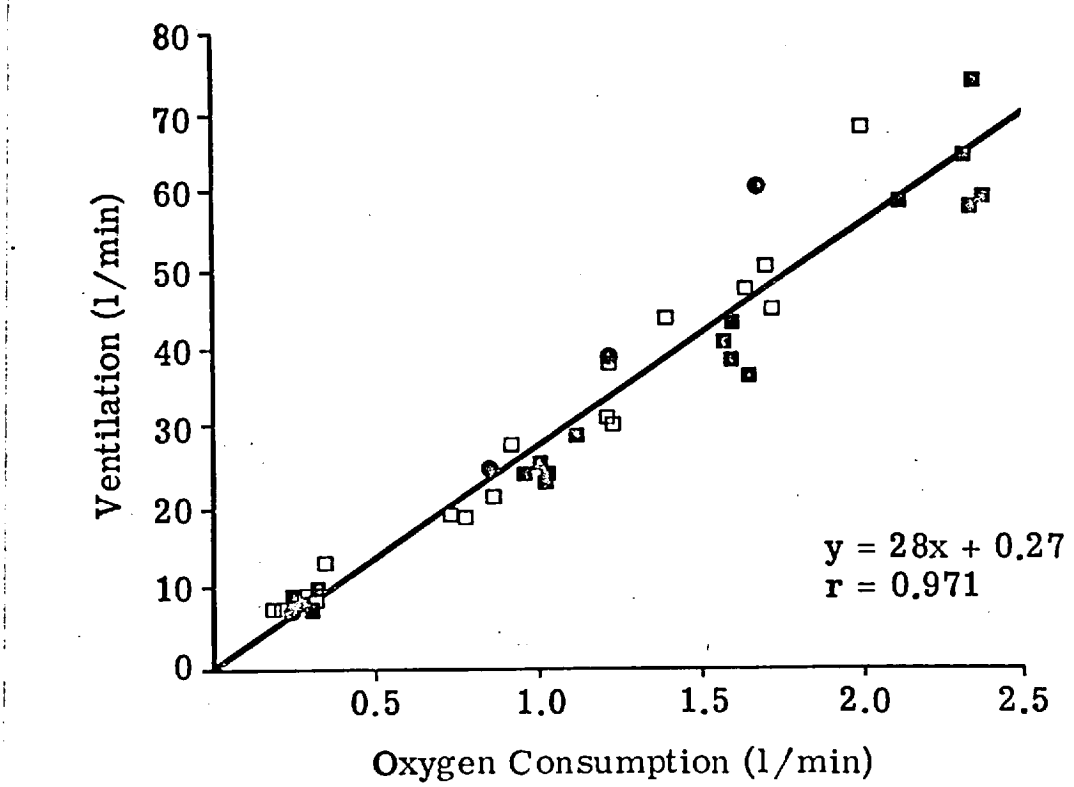

80
70
60
50
40
30
20
10
0
Ventilation (l/min)
0.5
1.0
1.5
2.0
2.5
Oxygen Consumption (l/min)
y = 28x + 0.27
r = 0.971

Figure 4.2

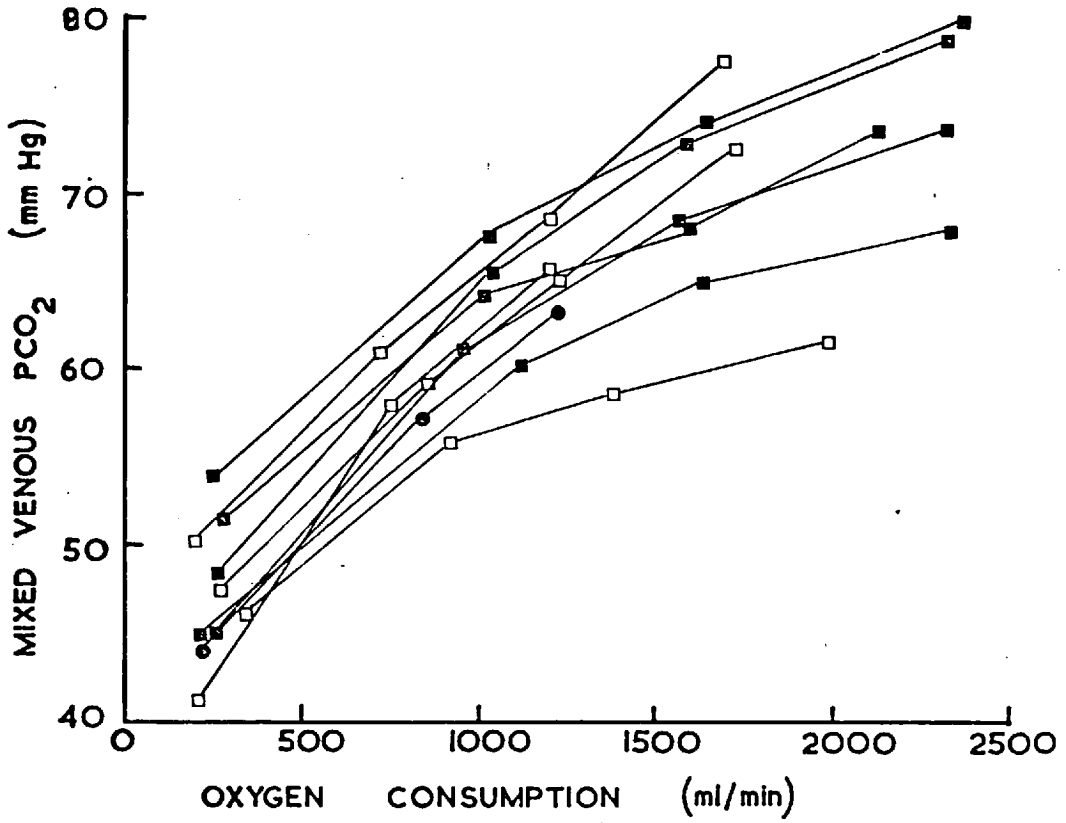

80
70
60
50
40
MIXED VENOUS PCO_2 (mm Hg)
0
500
1000
1500
2000
2500
OXYGEN CONSUMPTION (ml/min)

Figure 4.3

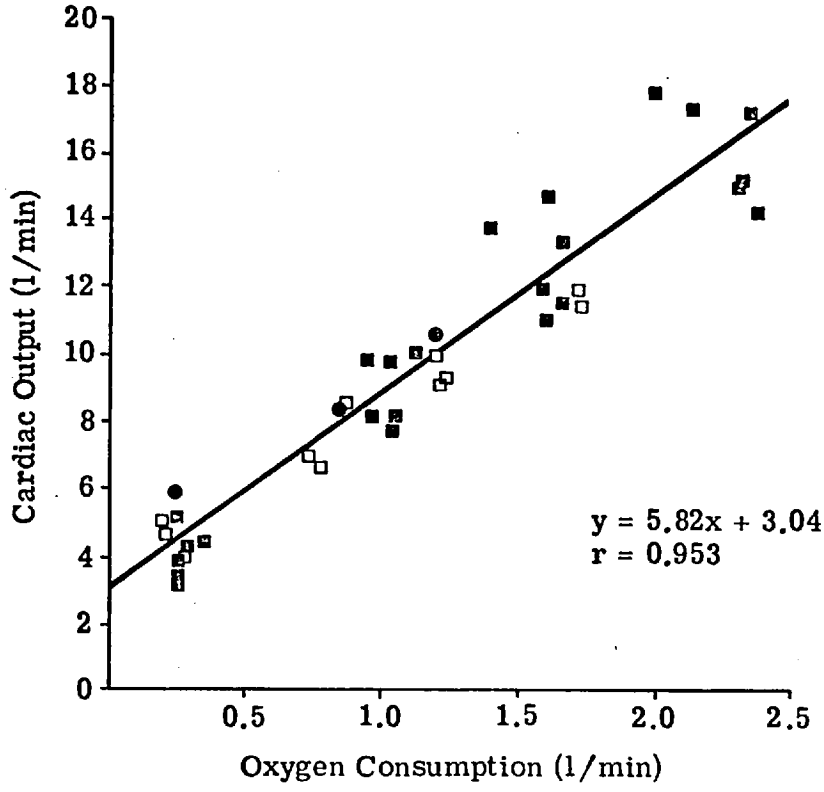

Cardiac Output (l/min)
20
18
16
14
12
10
8
6
4
2
0
0.5
1.0
1.5
2.0
2.5
Oxygen Consumption (l/min)
y = 5.82x + 3.04
r = 0.953

Figure 4.4

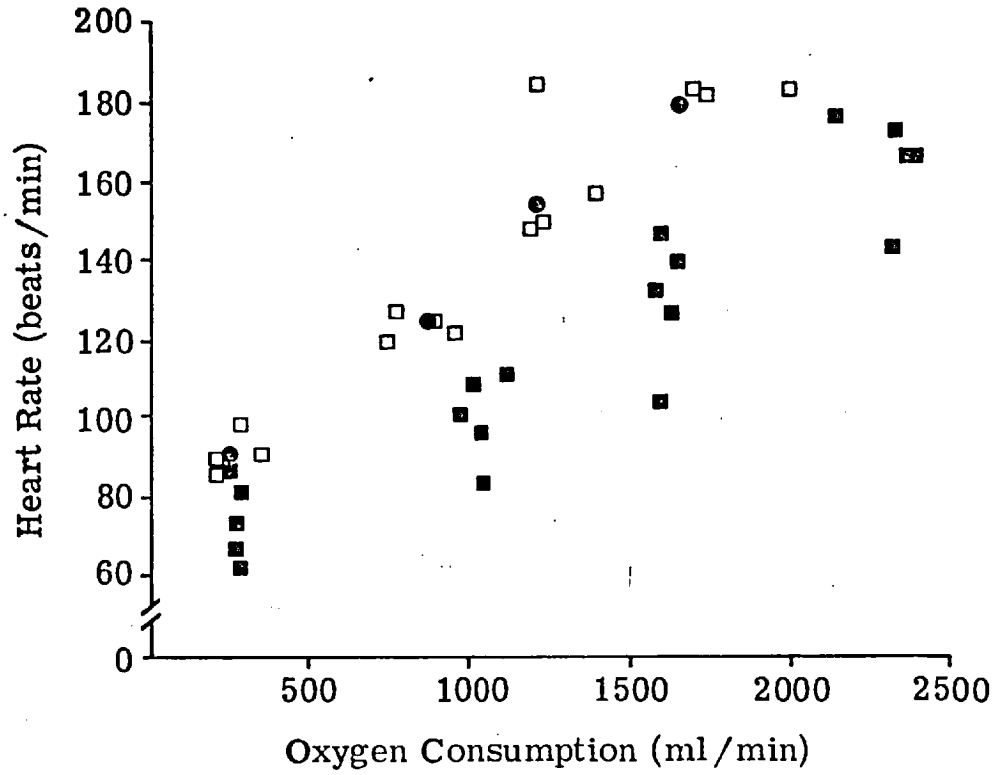

200
180
160
140
120
100
80
60
0
Heart Rate (beats/min)
500
1000
1500
2000
2500
Oxygen Consumption (ml/min)

Figure 4.5

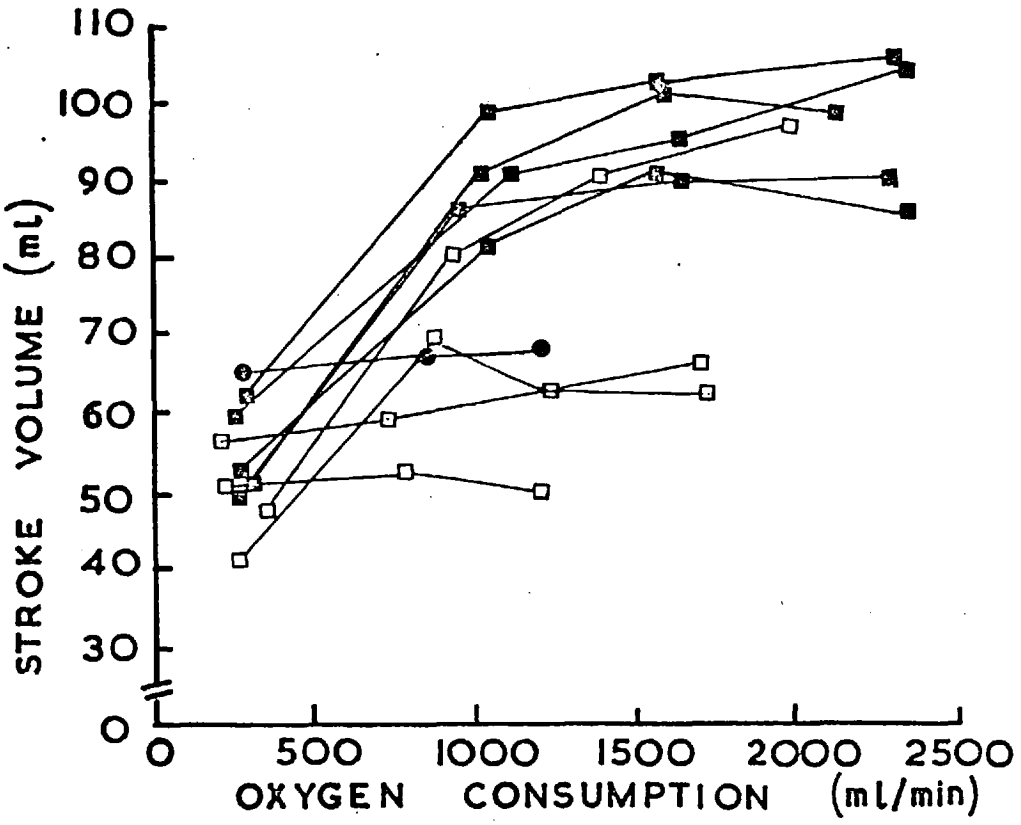

110
100
90
80
70
60
50
40
30
0
STROKE VOLUME (ml)
0
500
1000
1500
2000
2500
OXYGEN CONSUMPTION (ml/min)

Figure 4.6

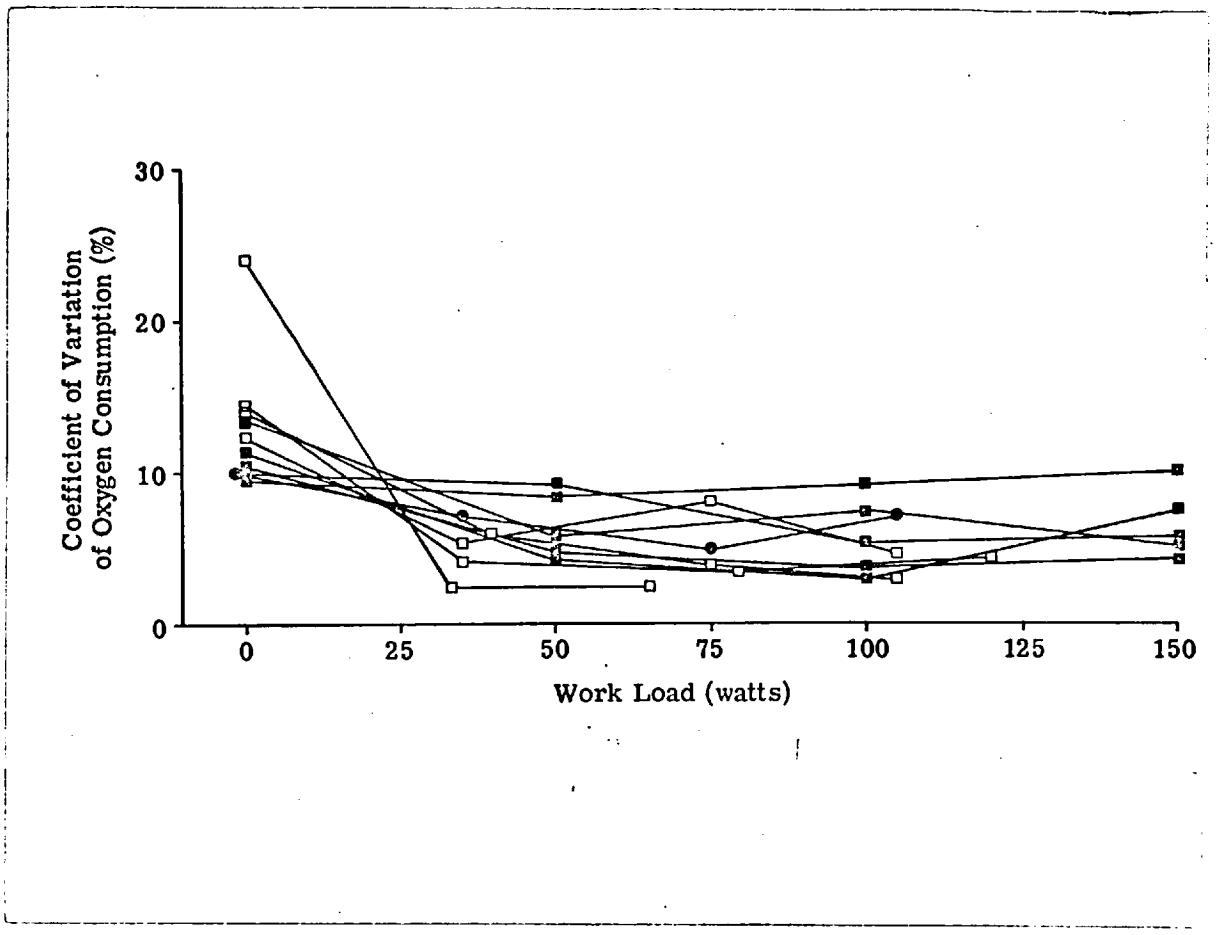


Figure 4.7 The coefficients of variation of oxygen consumption in ten subjects at rest and during exercise, plotted against corresponding work loads.

Figures 4.8-4.12 The coefficients of variation of ventilation (4.8), plateau $P\bar{v}CO_2$ (4.9), cardiac output (4.10), heart rate (4.11) and stroke volume (4.12) are plotted against corresponding mean values of oxygen consumption at several work loads in ten subjects.

Adult males ■; Adult female ●; Children □.

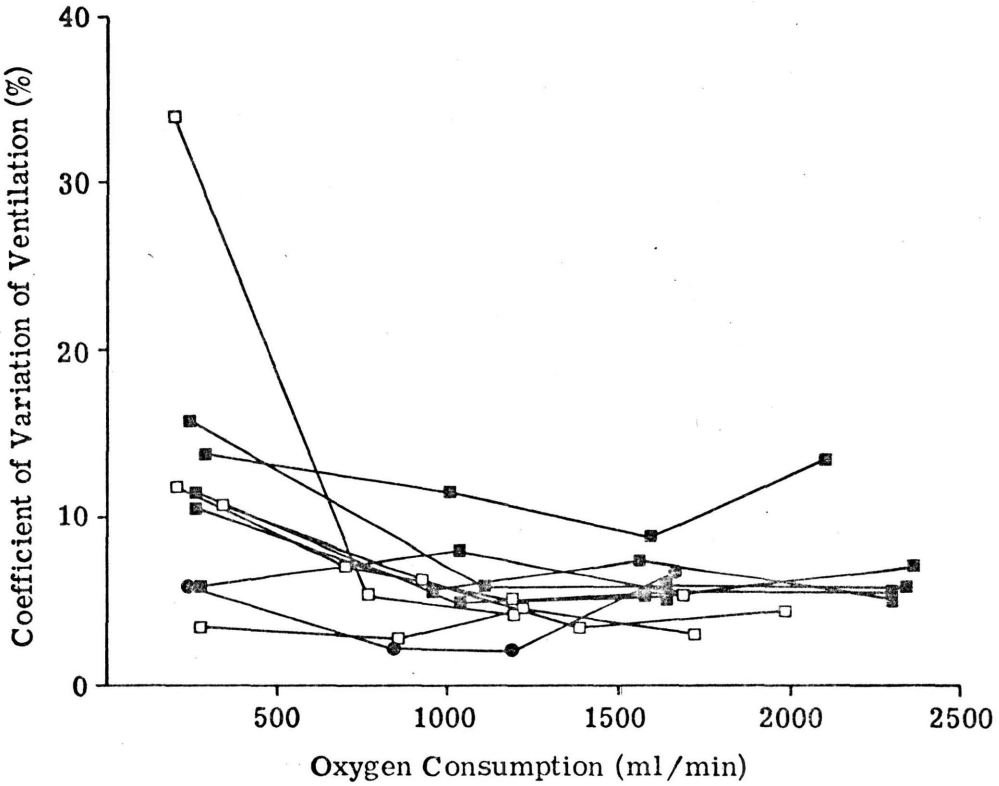

40
30
20
10
0
Coefficient of Variation of Ventilation (%)
500
1000
1500
2000
2500
Oxygen Consumption (ml/min)

Figure 4.8

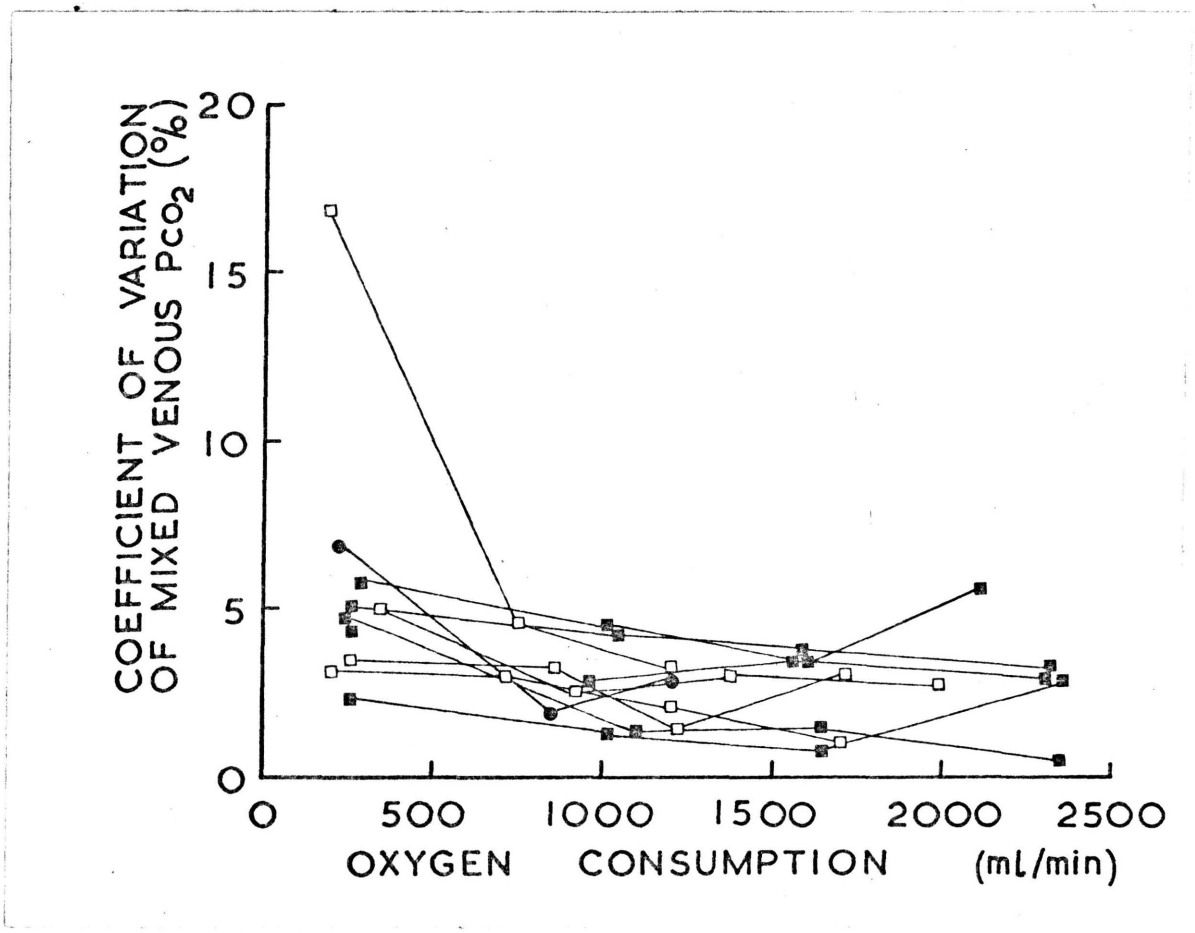

COEFFICIENT OF VARIATION
OF MIXED VENOUS P_{CO_2} (%)
20
15
10
5
0
0
500
1000
1500
2000
2500
OXYGEN CONSUMPTION (ml/min)

Figure 4.9

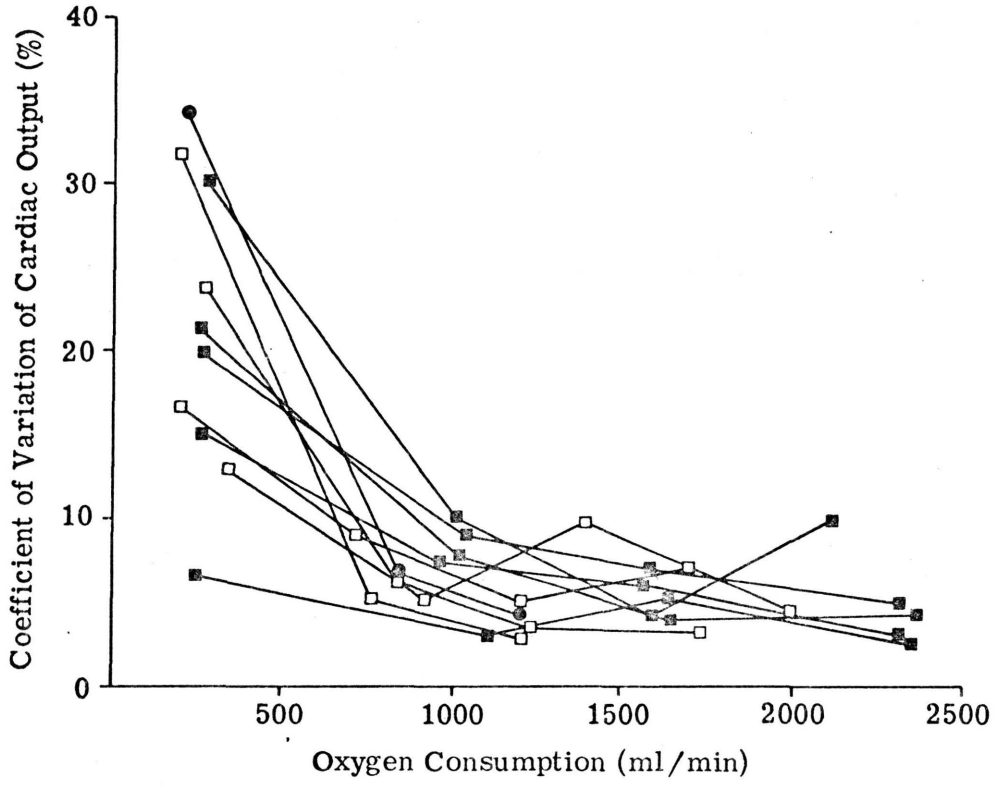

Coefficient of Variation of Cardiac Output (%)
40
30
20
10
0
500
1000
1500
2000
2500
Oxygen Consumption (ml/min)

Figure 4.10

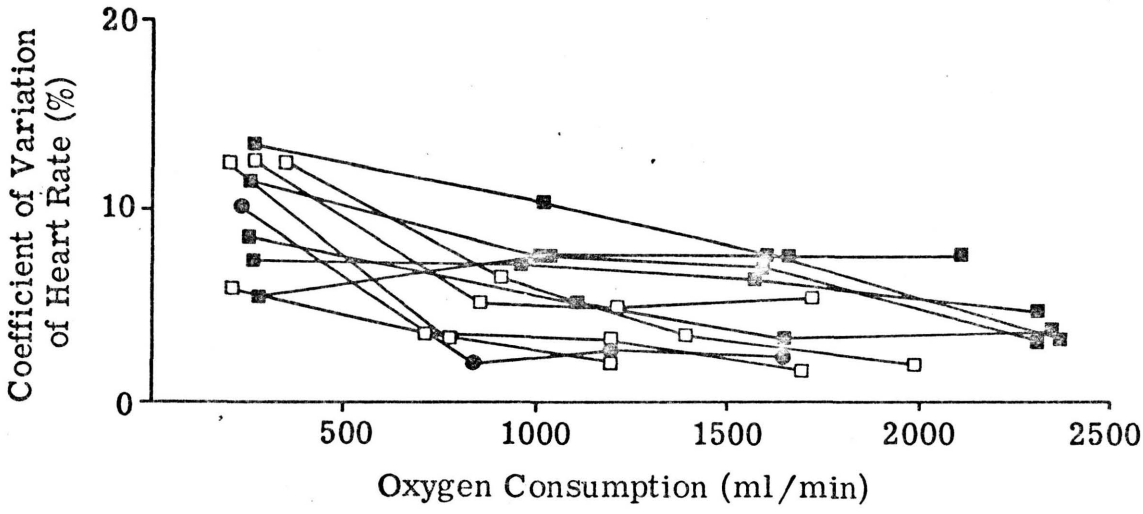

Coefficient of Variation of Heart Rate (%)
20
10
0
500
1000
1500
2000
2500
Oxygen Consumption (ml/min)

Figure 4.11

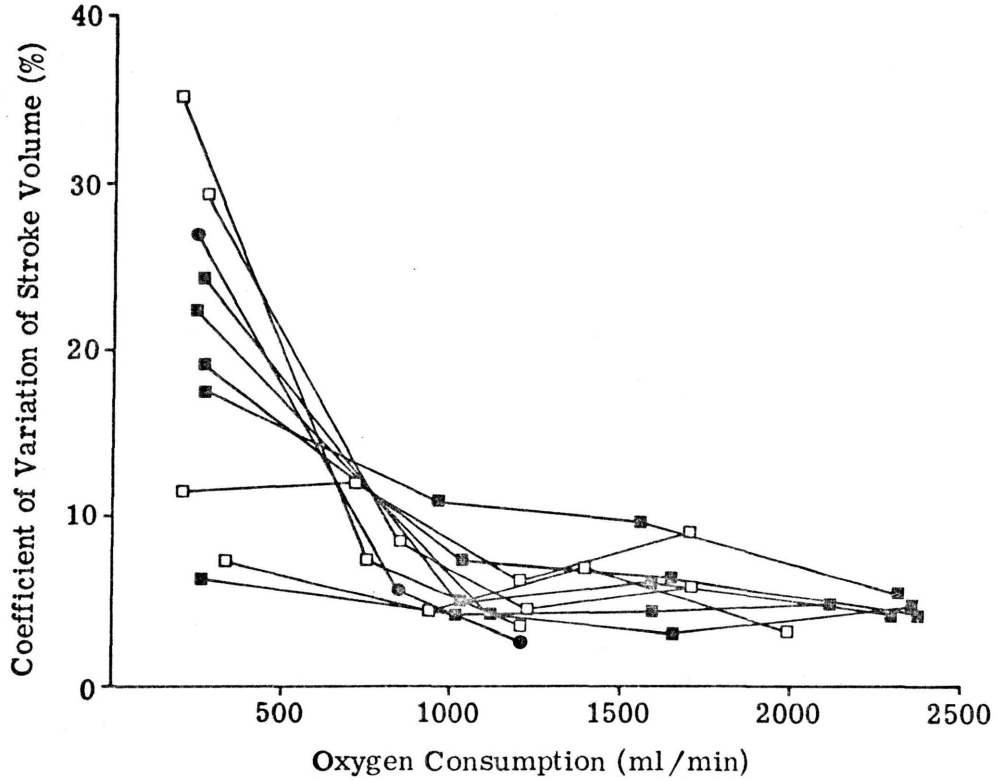

Coefficient of Variation of Stroke Volume (%)
40
30
20
10
0
500
1000
1500
2000
2500
Oxygen Consumption (ml/min)

Figure 4.12

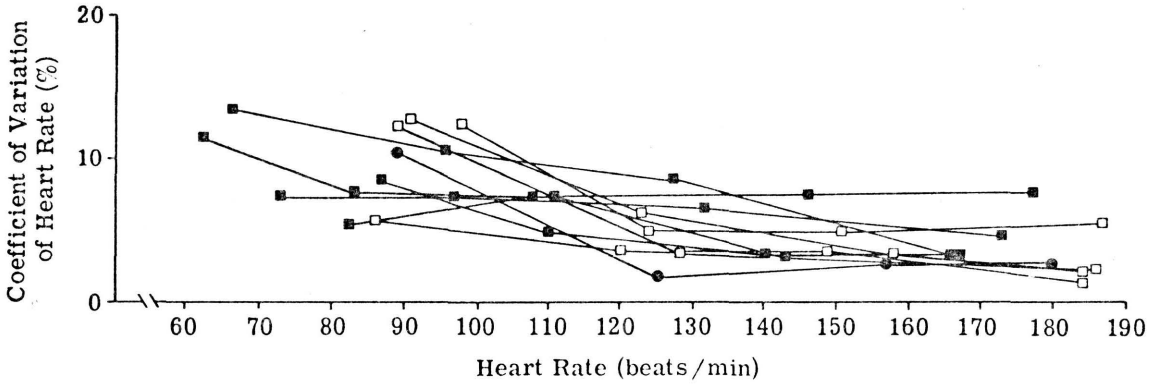


Figure 4.13 The coefficients of variation of heart rate measured in ten subjects at several work levels, plotted against the mean values of heart rate in each subject.

Table 4.2: The variability of cardiac output measured by the indirect (CO_2) Fick method. Reproducibility is assessed by the C.V.P.E. index.

OXYGEN CONSUMPTION (ml/min)	NUMBER of ESTIMATES (n) of CARDIAC OUTPUT	MEAN CARDIAC OUTPUT (l/min)	STANDARD DEVIATION of the DIFFERENCE (S.D. diff)	COEFFICIENT of VARIATION from PAIRED ESTIMATES (C.V.P.E.%)
252	20	4.18	0.56	9.5
927	20	8.13	0.50	4.4
1424	20	11.40	0.36	2.3
2108	16	16.10	0.50	2.4

DISCUSSION

The value of cardiac output and its reproducibility depends on the individual terms of the Fick equation. These terms include mixed venous PCO_2, arterial PCO_2, carbon dioxide output, and the slope of the CO_2 dissociation curve (discussed in Chapter 2).

THE MEASUREMENT OF $P\bar{v}CO_2$: The absolute values of $P\bar{v}CO_2$ were not related to the sex or age of the subjects, although there was considerable intersubject variation (Figure 4.3). The $P\bar{v}CO_2$ rose curvilinearly in all subjects, as carbon dioxide and oxygen consumption increased; the absolute estimate agreed with previously measured values in adults and children (Jones et al, 1967; Godfrey et al. 1971).

The day to day variation in the measurement of $P\bar{v}CO_2$ within subjects (7% at rest, and 2-3% during exercise) was generally as small as the variation in oxygen consumption, ventilation and heart rate. The latter were measured by simple methods which were independant of the rebreathing procedure, and where experimental error could be expected to be small. The variation in the measurements probably reflects true biological variation, and suggests that the variation in $P\bar{v}CO_2$ (which was of the same order) was also primarily biological.

The 2-3% coefficient of variation (C.V.) in daily measurements of $P\bar{v}CO_2$ was higher than that found by Godfrey and Wolf (1972). In their study, mixed venous PCO_2 was measured five consecutive times in five subjects who cycled for half an hour at work loads of

50 and 100 watts. They found that the mean coefficient of variation of $P\bar{v}CO_2$ within subjects was 1.4% and 0.8% at the two work loads. This may be taken as the minimum likely variation in the plateau rebreathing method due to technical and biological causes. In the present study, the small 1% variation in excess of this can probably be attributed to the day to day biological variation of mixed venous PCO_2.

THE MEASUREMENT OF ARTERIAL PCO_2: The arterial PCO_2 was calculated from the predicted normal physiological dead space in each subject. The values for dead space were taken from the study of Godfrey et al. (1971) who measured the PCO_2 of arterialised ear lobe blood to solve the Bohr equation for physiological dead space (Chapter 2). An error of $\pm$ 44 ml (95% confidence limits) could be made in estimating physiological dead space from their regression equation. However an advantage of predicting $PaCO_2$ from dead space was that large changes in the latter only altered the absolute values of $PaCO_2$ and cardiac output slightly (see later). Furthermore, any small errors in the prediction of dead space were recurrent in any one subject, thus having little or no effect on the reproducibility of cardiac output measurements.

THE VARIATION IN CARDIAC OUTPUT: At rest the mean coefficient of variation of cardiac output within subjects (using the indirect (CO_2) Fick equation) was 22%. The absence of real steady state conditions at rest was probably responsible for the high figure, as well as the high resting coefficients of variation of other terms in the Fick equation. For example, the mean coefficient

of variation (C.V.) of $\dot{V}CO_2$ within subjects was 15.5% (Table A-4.1); the coefficient of variation of $P\bar{v}CO_2$ was somewhat smaller (7%) only because changes in ventilation (C.V. 12.5%) have a mild effect on $P\bar{v}CO_2$, due to the large body stores of CO_2.

Another reason for the variability of cardiac output at rest is the small veno-arterial CO_2 pressure (or content) difference of blood. The pressure difference is of the order of 6-10 mm.Hg, and errors in estimating $P\bar{v}CC_2$ alone could alter this difference by 3 mm.Hg. This would cause an approximate 30-50% alteration in measured cardiac output.

During exercise there was a marked reduction in the variation of all parameters (Figures 4.6 - 4.13). The mean coefficient of variation of cardiac output within subjects was 2-10%. At an oxygen consumption of 1200 ml/min the mean C.V. of cardiac output within subjects was 5.7% and was comparable to the 3-6% C.V. found in the dye dilution and direct (O_2) Fick methods (Grimby, 1966; Ekelund and Holmgren, 1964). The greater reproducibility of the method during exercise is probably due to stabler steady state conditions, and to the enlarged blood veno-arterial CO_2 pressure or content differences, which is four or five times larger than the resting difference.

In order to compare the present results of cardiac output with some of the published data, its coefficient of variation was calculated from paired estimates (C.V.P.E.) in each of the ten subjects (Table 4.2), as described in the "statistics" section of

Chapter 2. At rest the C.V.P.E. for a single measurement of cardiac output was 9.5%, and was as good as the 8-20% variation found in the direct (O_2) Fick and dye dilution methods (Cournand et al., 1945; Werko et al. 1949; Richardson et al. 1959). During exercise the C.V.P.E. of cardiac output was between 2.2 - 4.4%, which was as good as, or better than the variation found by several workers using the direct Fick and dye dilution methods (Table 4.3). These workers have used the C.V.P.E. to assess the variation of their methods by taking consecutive paired estimates of cardiac output on a group of subjects. The C.V.P.E. of cardiac output in the present study compares very favourably with the published data (Table 4.3), despite the fact that the paired measurements were separated by at least one day.

COMPARISON OF ABSOLUTE VALUES OF CARDIAC OUTPUT: The cardiac output at a given oxygen consumption was similar in all subjects (Figure 4.4). When resting values of cardiac output were included, the regression line of cardiac output against oxygen consumption for the group was $\dot{Q}$ (l/min) = 3.04 + 5.80 $\dot{V}O_2$, = 0.95. However, excluding the resting values of cardiac output (where the mean C.V. of cardiac output was 22%), the regression equation was altered to $\dot{Q}$ (l/min) = 3.73 + 5.20 $\dot{V}O_2$. The regression line was similar to the regression lines of cardiac output from several studies in which invasive and non-invasive methods were used. The data from several authors are summarised in Table 4.4. The table also includes data collected by the author from several studies in the literature (Table A-8.1).

Table 4.3: The variability (measured as C.V.P.E.%) of various methods of measuring cardiac output during exercise.

SOURCE	METHOD of MEASURING CARDIAC OUTPUT	COEFFICIENT of VARIATION of PAIRED ESTIMATES of CARDIAC OUTPUT (C.V.P.E.%) Range during exercise
Present Study	INDIRECT FICK (CO_2 REBREATHING) METHOD	2.3 - 4.4 %
Holmgren & Pernow, 1960 Clausen, et al. 1970	DIRECT (O_2) FICK METHOD	5.6 - 7.5 %
Clausen, et al. 1970	DIRECT (CO_2) FICK METHOD	8.2 %
Clausen, et al. 1970 Ekblom, 1969 Hanson & Tabakin, 1964	DYE DILUTION METHOD	3.5 - 10 %

Table 4.4: Values of cardiac output for normal subjects, quoted by workers using the dye dilution, direct (O_2) Fick and the indirect (CO_2) Fick methods during exercise. Values of cardiac output at 1 and 2 l/min oxygen consumption were derivedfrom regression equations.

SOURCE*	No. of observations (n)	SUBJECTS	METHOD	CARDIAC OUTPUT (l/min) at an OXYGEN CONSUMPTION of	
				1 l/min	2 l/min
Present study	254	Adults and children	$\dot{Q}CO_2$ uncorrected	8.9	14.1
Zeidifard (chapter 7)	102	Adults	$\dot{Q}CO_2$ uncorrected	10.0	14.8
Godfrey et al. (1971)	161	Children	$\dot{Q}CO_2$ uncorrected	9.6	15.2
Eriksson et al. (1971)	8	Children	DYE dilution	9.8	14.4
Collected data (Zeidifard)	209	Adults	DYE dilution	11.1	15.8
Collected data (Zeidifard)	56	Adults	FICK O_2	10.8	16.6
Edwards et al. (1969)	12	Adults	$\dot{Q}CO_2$ corrected	12.8	19.4
Denison et al. (1971)	60	Adults	$\dot{Q}CO_2$ corrected	14.9	22.6
Spiro et al. (unpublished)	30	Adults	$\dot{Q}CO_2$ corrected	11.5	17.7
Zeidifard (chapter 7)	102	Adults	$\dot{Q}CO_2$ corrected	12.3	18.3

* The sources of collected data are presented in Table A8.1.

Although the slope of the present regression line (5.20 l/ litre oxygen consumption) was similar to the slope found by other workers, the absolute values of cardiac output were slightly lower.

Several factors which may have caused this were considered. Training is known to cause a diminution in ventilation, heart rate and cardiac output (Davies, et al. 1970; Andrews et al. 1966). However in this study, an analysis of covariance showed no consistent reduction in these parameters. Similarly, although low cardiac outputs have been reported for children (Eriksson et al. 1971; Godfrey et al. 1971) no significant differences were found between the adults and children in this study.

It is more likely that a slight underestimation of dead space (and arterial PCO_2) in the adult subjects caused the low values of cardiac output. The few studies which have measured dead space in exercising adult subjects (Asmussen and Neilsen, 1952; Jones et al., 1966) have assumed a mean value of dead space for all subjects at rest and have suggested an increase of 7-8 ml dead space for a 100 ml increase in tidal volume. This study used the more comprehensive data of Godfrey et al. (1971) on adolescents and children, which related dead space to body weight and found a smaller increase of 5 ml dead space for every 100 ml. increase in tidal volume. The study also gave confidence limits for the estimation of dead space ($\pm$ 44 ml.), and therefore of arterial PCO_2. The overall under-estimation of dead space using this data (instead of the data of Asmussen and Neilsen, 1952; and Jones et al. 1966) would have under-estimated cardiac output by about 0.8 l/min.

Finally the present value of cardiac output could have been raised if a small correction had been applied for the difference between gas and blood $\bar{P}CO_2$ during the plateau rebreathing procedure, which has been observed in both adults and children (Jones et al. 1967 and 1969; Denison et al. 1971; Godfrey et al. 1971b). However, values of cardiac output based on the uncorrected estimate of $\bar{P}vCO_2$ compare favourably with those found simultaneously by the direct (O_2) Fick method, and also with values in the literature; in comparison, corrected values are often 2-3 litres higher (Denison et al. 1969; Edwards et al. 1969; Godfrey et al. 1971; Table 2; see also Chapter 7).

Although the cardiac output values in this study were low, they were only marginally so (by about 1 l/min), and corresponded well with values measured by direct invasive techniques. The measurements of stroke volume (Figure 4.6) also suggest that cardiac output was measured reliably, since the values were similar to those found by Bevegard et al. (1960) and Godfrey et al. (1971), and were appropriate to the size and sex of the subjects.

CONCLUSIONS:

The reproducibility of cardiac output measured by the indirect (CO_2) Fick method was found to be as good as that of the direct (O_2) Fick and dye dilution methods. The absolute values of cardiac output also compared well with those derived from invasive techniques. In common with these methods the indirect (CO_2) Fick method was unreliable at rest, but became extremely reproducible at all levels of work. The inaccuracy of the method at rest is probably due to the small veno-arterial CO_2 content difference of the blood, and to the difficulty of obtaining steady state conditions.

During exercise both these limitations are removed to allow a more accurate calculation of cardiac output.

CHAPTER 5

A COMPARISON OF CARDIAC OUTPUT MEASURED BY THE AYOTTE AND THE MODIFIED CANDER & FORSTER N_2O REBREATHING METHODS

INTRODUCTION

There are several rebreathing methods which use a foreign gas such as acetylene or nitrous oxide to measure cardiac output by non-invasive means. Most of the methods are identical in use, but differ in the interpretation of relevant data. Several methods have been discussed in the Introduction (Chapter 1), while those which are examined in the present study are discussed in Chapter 2 (Section b). The methods investigated in this chapter are those described by Ayotte et al. (1970), and by Cander and Forster (1959); for the purposes of this work the latter was modified from a breathholding to a rebreathing technique. A new method which was developed at a later stage is described in Chapter 6.

SUBJECTS & METHODS

Seven normal subjects volunteered for the study: some of their anthrompometric data are presented in Table 5.1. Each subject exercised on a bicycle ergometer for periods of half an hour each day at work loads of 40, 80 or 120 watts. In the first five minutes of the test, heart rate, ventilation and gas exchange were monitored continuously until a steady state of exercise was obtained. Mixed venous PCO_2 was then estimated by the plateau CO_2 rebreathing technique in order to calculate the cardiac output by the indirect (CO_2) Fick method. At the end of the rebreathing procedure, normal breathing was resumed for a further five minute period. The subject then performed a second rebreathing manouvre (10 seconds duration) with a 4 litre mixture of

Table 5.1: Anthropometric data of the subjects examined in the study.

SUBJECT	AGE (yrs)	SEX	Ht (cm)	Wt (kg)
PF	33	M	178	73.5
GP	25	M	175	72.0
SS	23	M	183	80.6
MR	30	M	180	75.5
JP	28	M	187	73.2
JJ	28	M	176	76.0
PK	34	M	164	62.5

8% nitrous oxide, and 0.5% neon in air. An infra-red N_2O analyser was used to monitor the alveolar N_2O concentration during the rebreathe, as well as the concentrations of N_2O in the bag before and after each rebreathe manouvre. 40 ml samples of the initial and final bag mixture were collected in syringes, and were then analysed on a gas chromatograph for their neon gas content.

Over the half hour period of exercise each subject performed a total of one CO_2 and four N_2O rebreathing procedures which were each carried out at intervals of 5-6 minutes.

CALCULATIONS: The analysis of the results is detailed in Chapter 2. Cardiac outputs based on the Ayotte (1970) and modified Cander and Forster (1959) methods were both calculated from the same N_2O rebreathing manouvre. The total lung-bag rebreathing volume was calculated from the dilution of the inert neon gas. In the Ayotte method, the uptake of nitrous oxide was calculated from a difference in the initial and final amounts of N_2O in the rebreathing bag, corrected for a small amount of N_2O dissolved in lung parenchymal tissue. The pulmonary blood flow was obtained by dividing the uptake of nitrous oxide by the true mean alveolar concentration of N_2O during the rebreathing period (Chapter 2: Section b). This was calculated from the planimetered area underneath a rebreathing trace (Figure 2.10), in units of N_2O atmosphere-seconds.

In the modified Cander and Forster method, the uptake of nitrous oxide was measured from the exponential loss of N_2O from the system, determined from a semilogarithmic plot of the end-expired N_2O

concentration during the rebreathing period. This amount was corrected for N_2O dissolved in lung tissue, as before. The exponential form of the uptake of nitrous oxide predicts that in a fixed lung-bag rebreathing volume, the change in concentration of N_2O at any instant is directly related to both the initial concentration at that time, and to the blood flow which is removing the gas. The change in concentration over a period of time thus becomes an exponential function of the blood flow which is then calculated. Details of this procedure are discussed in Chapter 2 (section b).

RESULTS

The results of the study are summarised in Table A5 - 1 of the appendix. For each subject values of oxygen consumption, heart rate, and cardiac output (CO_2 rebreathing method) are shown for various work loads. Subject J.J. did not perform the highest work load, while subject G.P. did not complete it.

Four consecutive estimates of cardiac output based on the Ayotte and modified Cander and Forster N_2O rebreathing methods are presented for each subject; mean values and their coefficient of variation are also given. The mean values of cardiac output determined by the Cander and Forster N_2O method in each subject, are plotted against oxygen consumption in Figure 5.1. Two regression lines are plotted in the figure, and represent the group values of cardiac output based on the Cander and Forster N_2O and the CO_2 rebreathing methods. Estimates of cardiac output based on the Ayotte method are similarly presented in Figure 5.2.

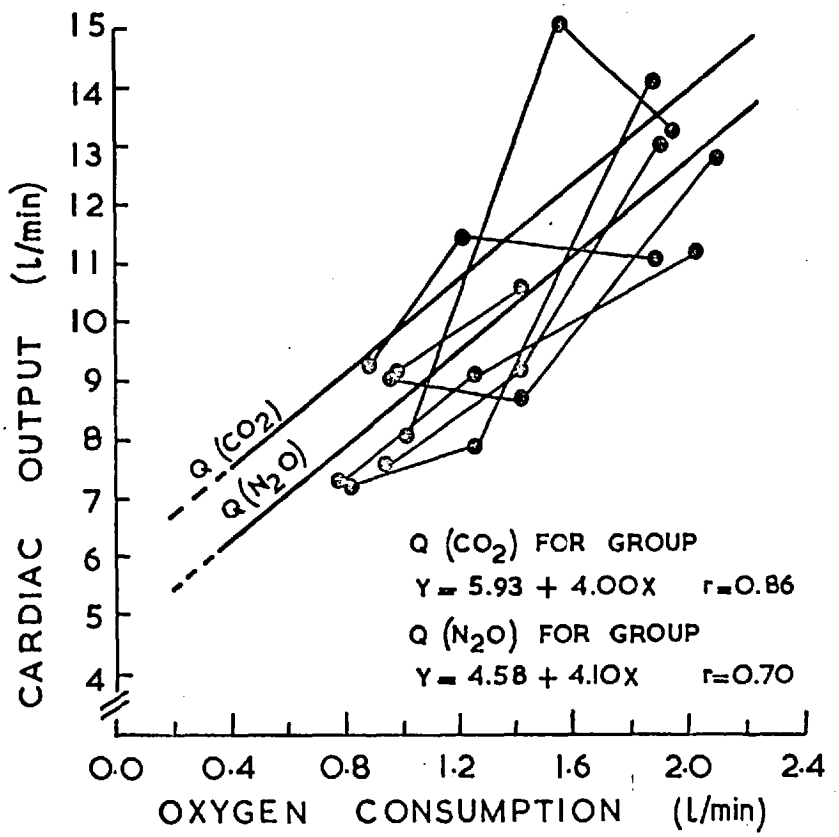


Figure 5.1 Mean values of cardiac output measured by the Cander and Forster N_2O rebreathing method are plotted against the oxygen consumptions at three work levels in seven subjects. A regression line for the group data is also presented. A second regression line, based on the indirect (CO_2) Fick estimates of cardiac output, is shown for comparison.

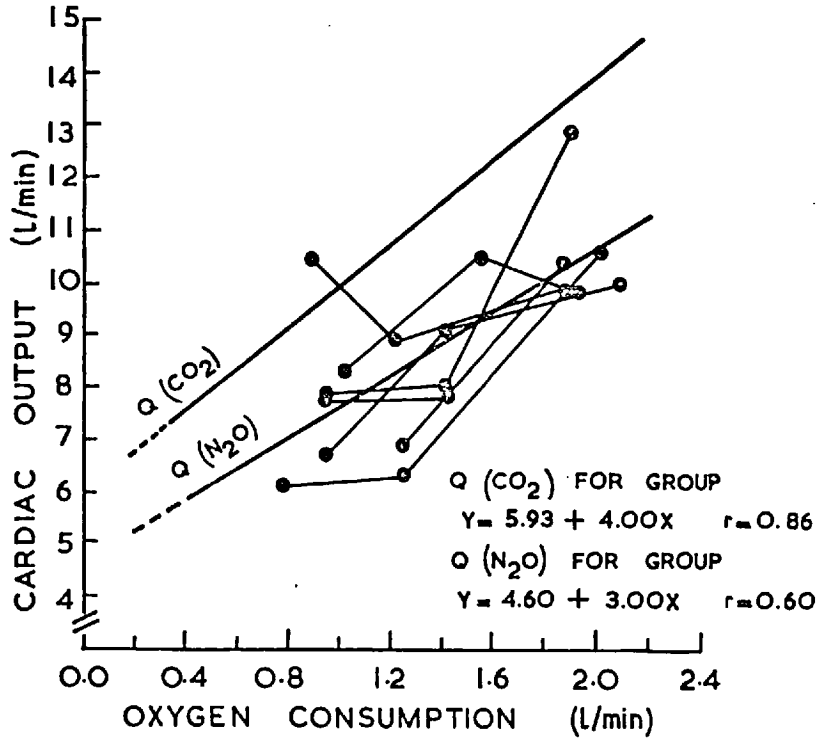


Figure 5.2 Mean values of cardiac output measured by the Ayotte et al. N_2O rebreathing method are plotted against the oxygen consumption at three work levels in seven subjects. A regression line for the group data is also presented. A second regression line, based on the indirect (CO_2) Fick estimates of cardiac output is shown for comparison.

Both the N_2O rebreathing methods gave low values of cardiac output in comparison with the CO_2 rebreathing method. The Ayotte estimates of cardiac output ($\dot{Q}$ Ayotte) were generally lower than the values measured by the Cander and Forster method ($\dot{Q}$ Cander). In addition both methods showed considerable variation for consecutive estimates of cardiac output in individual subjects. This was clearly demonstrated when the coefficients of variation of cardiac output (C.V.) within subjects were plotted against oxygen consumption in Figures 5.3 and 5.4. The coefficients of variation of paired estimates of cardiac output (C.V.P.E.) were also high in both methods (6-13%; Table 5.2).

DISCUSSION

The same data was used to calculate cardiac output by the Ayotte and the modified Cander and Forster methods, so that instrumentation errors and the biological variation of the subjects were identical in both cases.

Relative to the indirect (CO_2) Fick estimates of cardiac output, those from both N_2O rebreathing methods were low (Figures 5.1 and 5.2). Furthermore, values based on the Ayotte method were generally lower than those calculated by the modified Cander and Forster method. For both methods the variability of consecutive estimates of cardiac output in individual subjects was large (Figures 5.3 and 5.4), however for any one subject the variability in these methods was often markedly dissimilar.

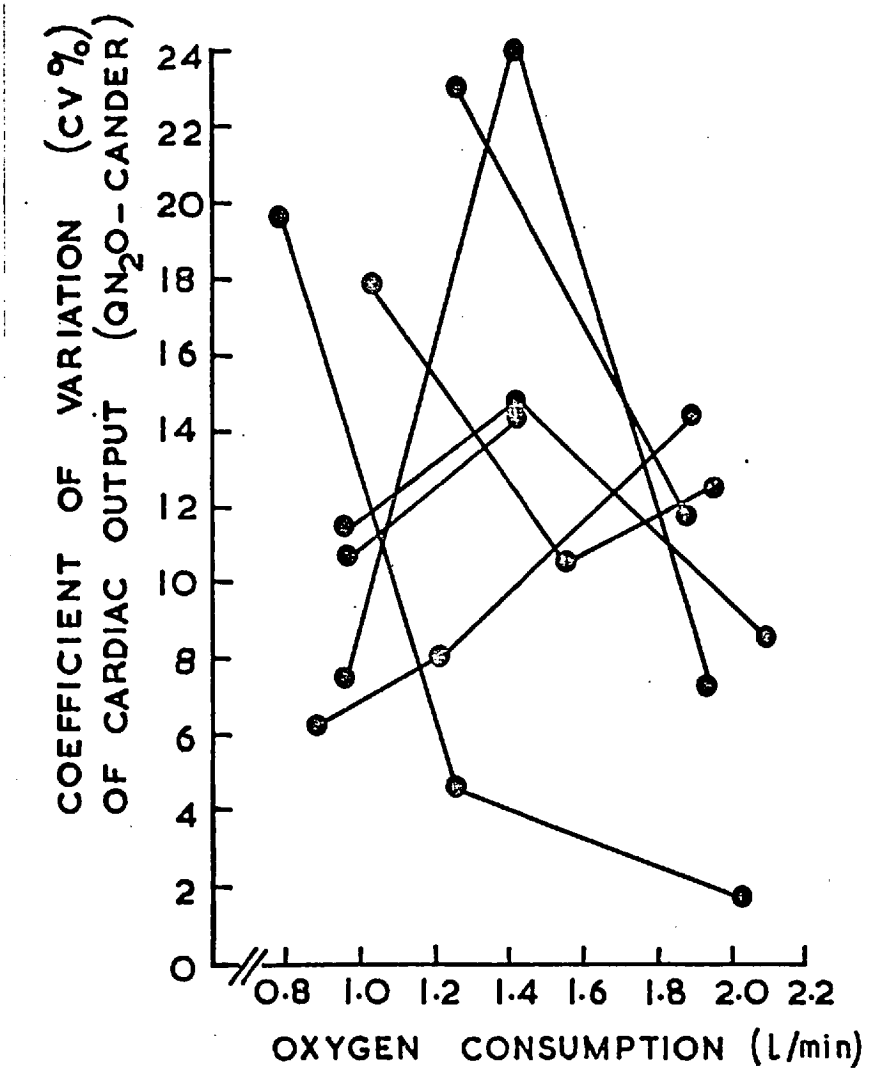


Figure 5.3 The mean coefficients of variation of cardiac output determined by the modified Cander and Forster N_2O rebreathing method during exercise, are plotted against the oxygen consumptions at each work load in seven subjects.

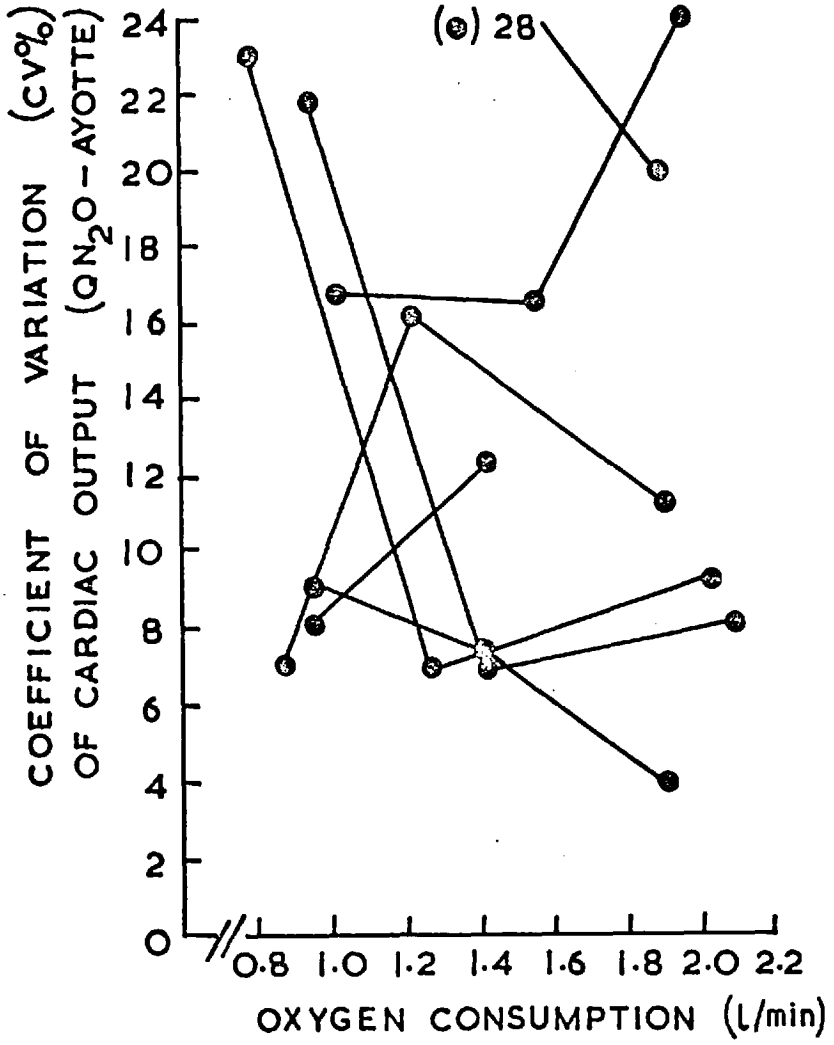


Figure 5.4 The mean coefficients of variation of cardiac output determined by the Ayotte et al. N_2O rebreathing method during exercise, are plotted against the oxygen consumptions at each work load in seven subjects.

Table 5.2: A comparison of the variability of the N_2O rebreathing methods with that of other methods of measuring cardiac output.

SOURCE	METHOD of MEASURING CARDIAC OUTPUT	COEFFICIENT of VARIATION of PAIRED ESTIMATES of CARDIAC OUTPUT (C.V.P.E.%) Range during exercise
Present Study	N_2O REBREATHING METHOD of AYOTTE	7.2 - 13.2 %
	MODIFIED N_2O REBREATHING METHOD of CANDER and FORSTER	6.5 - 10.2 %
Holmgren & Pernow, 1960	DIRECT (O_2) FICK METHOD	5.6 %
Ekblom, 1969	DYE DILUTION METHOD	3.0 - 3.5 %
Zeidifard (chapter 4)	INDIRECT (CO_2) FICK METHOD	2.3 - 4.4 %

VARIABILITY OF THE METHOD: The high coefficients of variation of cardiac output (measured as C.V. or C.V.P.E.) indicate a large inconsistency in the measurement of cardiac output by both methods (Figures 5.3, 5.4 and Table 5.2). The overall coefficients of variation (C.V.) are as high as 23%, in marked contrast to a 3-5% coefficient of variation in cardiac output measured by the indirect (CO_2) Fick, direct (O_2) Fick and dye dilution methods (Chapter 4; Grimby, 1966; Ekelund and Holmgren, 1964). The 6-13% coefficients of variation of paired estimates of cardiac output (C.V.P.E.) were also high in contrast with the 2 - 7% C.V.P.E. of cardiac output using invasive techniques (Table 5.2).

To investigate the large variations in the measured cardiac output, potential errors in various parameters contributing to its value were examined. Sources of error included the calculation of:

1) lung-bag rebreathing volume
2) lung tissue volume
3) nitrous oxide uptake
4) mean alveolar concentration of nitrous oxide

CALCULATIONS OF LUNG-BAG VOLUME: The uptake of nitrous oxide was calculated from its loss from the lung-bag rebreathing volume. This volume was assumed to remain unchanged throughout the rebreathing manouvre, so that the drop in the N_2O concentration became an exponential function of blood flow and the fixed rebreathing volume. The assumption of a fixed volume could only be true if the removal of nitrous

oxide and oxygen were balanced by addition of carbon dioxide into the lung-bag system. However, in the rebreathing procedure, an exponential release of carbon dioxide was balanced against an exponential uptake of nitrous oxide and a continual uptake of oxygen, so that the exchange of gases produced a net removal of gas from the rebreathing system. The assumption is therefore wrong, and its significance would depend on the size of the rebreathing volume and the initial concentration of nitrous oxide in the rebreathing bag. The small changes in total rebreathing volume (due to an imbalance of gas exchange) would have a less pronounced effect on a large rather than a small volume. However the upper limit of the rebreathing volume is dictated by the requirement of complete lung-bag mixing within a period of about four seconds. The initial concentration of N_2O would also affect the degree of error, since higher concentrations cause a greater uptake of gas, and create a correspondingly larger imbalance in gas exchange. An imbalance in the exchange could also be produced by the increased oxygen uptake at high work loads.

Using a modified rebreathing procedure described in Chapter 6 it was found that the cardiac output at an oxygen consumption of 2 l/min ($\dot{Q}$ = 15.7 l/min) was under-estimated by 35% if changes in total rebreathing volume (which were comparable to those in this study) were not taken into account.

THE MEASUREMENT OF TISSUE VOLUME: In both the Ayotte and modified Cander and Forster methods an allowance was made for a volume of

nitrous oxide dissolved in lung tissue. Its omission would have caused a 6 - 9% under estimation in cardiac output (Cander and Forster, 1959); however, the error would have been systematic.

THE UPTAKE OF NITROUS OXIDE: In the modified Cander and Forster method errors in the measured change in N_2O concentration during the rebreathe were minimised by plotting the end-expired N_2O concentration with each breath. However, since the assumption of a fixed total rebreathing volume was incorrect it was also not strictly correct to assume an exponential rate of loss of nitrous oxide into pulmonary blood. The degree of underestimation of cardiac output due to the assumption has already been discussed. It is notable that even at the low work loads examined by Johnson et al. (1960), the values of pulmonary blood flow (given in ml/min x M2) shown in Figure 7 of their paper were considerably lower than those of Riley et al. (1948) and Donald et al. (1948).

In the Ayotte method the loss of N_2O was determined from the initial and final mixed bag concentrations of nitrous oxide. Apart from the practical undesirability of such a procedure, a true variation in the final bag N_2O concentration could easily be produced by the effort dependant procedure of exhaling to residual volume. This variation would have been reflected in unsystematic errors in the measurement of cardiac output.

THE MEAN ALVEOLAR N_2O CONCENTRATION: The most likely cause of the low values of cardiac output in the Ayotte method (relative to those from the Cander and Forster method) lay in the measurement

of the mean alveolar N_2O concentration. This was calculated from the planimetered area underneath the rebreathing trace, to give a time integrated alveolar fraction of N_2O in units of atmosphere-seconds. The technical error of planimetry was less than 1%; however a major theoretical error was made in assuming that the planimetered area comprised exclusively of the alveolar gas fraction. In the first few seconds of the rebreathe, the tidal concentration of N_2O depended on the mixing of N_2O between lung and bag volumes, and was in no way related to the alveolar N_2O concentration. Its planimetry would generally have over-estimated the mean alveolar N_2O concentration, with a resultant under-estimation of cardiac output in the Ayotte method. However, in the study of Ayotte et al. (1973) the regression line of cardiac output appears normal, although there is a wide distribution of individual values (Figure 5.5). In a previous study (Ayotte et al. 1970) their individual values of cardiac output were high compared to those found in the literature, and gave a minimum value of 8 l/min for cardiac output at rest. The discrepancy between their observations and those of this study cannot be explained simply. In common with the present work, the authors used an infra-red N_2O analyser which was basically alinear, being less sensitive to high N_2O mixtures in the 0 - 10% working range. However Ayotte and his colleagues used a theoretical correction for the alinearity to calculate a true planimetered mean alveolar N_2O concentration. If their correction under-estimated the alinearity of their instrument, it would have normalised or increased their measured cardiac output (Figure 5.5), leaving the reproducibility of the method unaffected.

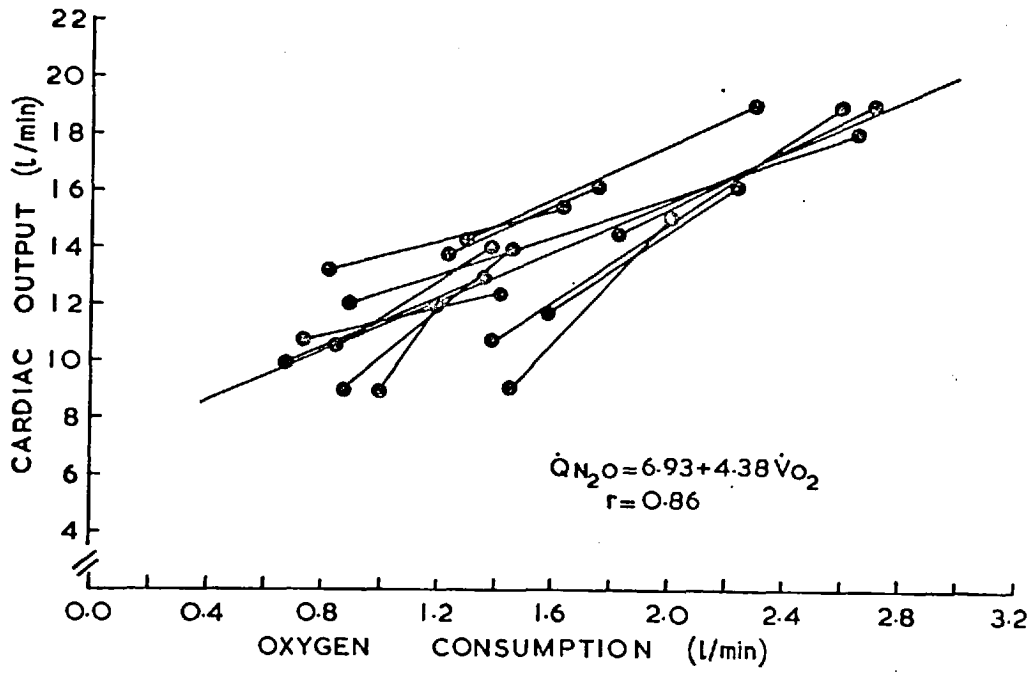


Figure 5.5 Cardiac output measured by the Ayotte et al. (1970) N_2O method in 13 normal subjects during exercise, plotted against corresponding values of oxygen consumption; a regression line for the data is also presented. The graph is compiled from the data of Ayotte et al, 1974.

Unfortunately it is difficult to comment on this aspect of Ayotte et al.'s studies since there is very little reproducibility data for their method. Two studies which have been published each quote a single value for the coefficient of variation of cardiac output within subjects. A value of 7.1% was given in Ayotte et al., 1970, and this was raised to 13.4% in Ayotte et al., 1973. Unfortunately, in the latter study although duplicate estimates of cardiac output were made on each subject, these were not presented in the paper and were not treated to statistical analysis. The two quoted values of the coefficient of variation in these papers were both within the range found in this study (Figure 5.4).

CONCLUSIONS

The values of cardiac output measured by the Ayotte and modified Cander and Forster N_2O rebreathing methods were lower than expected in normal subjects. The reproducibility of these methods was also poor and the most likely sources of error were the assumptions which were made when cardiac output was calculated.

The assumption of a fixed lung-bag rebreathing volume was probably responsible for the gross under-estimation of cardiac output in both methods.

The error in measuring the mean alveolar N_2O concentration by planimetry was probably the cause of the lower values of cardiac output in the Ayotte method.

These sources of error, with others which have been discussed, were probably also responsible for the poor reproducibility of the two methods.

Several modifications to the N_2O rebreathing method were indicated from the present study. In order to measure the cardiac output correctly, it was necessary that -

1. Changes in lung-bag volume during the rebreathing period should be calculated from the continuous measurement of an inert tracer gas.

2. As before, the concentration of N_2O should be monitored throughout a rebreathing period; however, the mean alveolar N_2O concentration should be calculated after the initial period of lung-bag gas mixing.

3. To eliminate any sources of technical error in data retrieval, it was thought desirable that all measurements during the rebreathing period were made with as few pieces of equipment as possible.

CHAPTER 6

A MODIFIED NITROUS OXIDE REBREATHING METHOD FOR THE MEASUREMENT OF CARDIAC OUTPUT

INTRODUCTION

In an examination of the Ayotte (1970) and the modified Cander and Forster (1959) N_2O rebreathing methods, it was found that both methods contained faults which affected the reproducibility and accuracy of the measured cardiac output (Chapter 5). The chief source of error was an under-estimation of cardiac output due to the assumption of an unchanged lung-bag volume during the rebreathing period. In the Ayotte method there were further potential errors due to the calculation of the mean alveolar N_2O concentration during the rebreathing period.

A review of the foreign gas breatholding and rebreathing methods as used between 1928 and 1973 suggested four main precautions which were necessary for an accurate measurement of cardiac output.

1. In terms of a rebreathing procedure, the method must ensure that the foreign gas in the bag and lung volumes is fully mixed before uptake of the gas is measured.

2. Changes in the total rebreathing volume during the rebreathing period must be taken into account.

3. The uptake of gas should not be estimated after significant recirculation of pulmonary blood, since it is diminished by the reduced partial pressure gradient of the gas between the alveoli and pulmonary blood.

Table 6.1: A survey of the literature on foreign gas breatholding and rebreathing methods. Each study was examined to determine whether:

(i) cardiac output was measured prior to recirculation of pulmonary blood.
(ii) the observed uptake of foreign gas was corrected for change in breatholding or rebreathing volume.
(iii) an allowance was made for the absorption of gas by lung tissue.

A tick indicates a correction was applied, while a cross indicates its omission.

Authors	Before Recirculation	Changes in lung-bag volume	Lung tissue volume
Grollman 1929	X	✓	X
Christensen 1931, (a & b)	X	✓	X
Chapman et al., 1950	X	✓	X
Asmussen & Neilsen 1952	✓	✓	X
Cander & Forster 1959	✓	X	✓
Saltin 1964	✓	✓	X
Rigatto 1967	✓	✓	X
Ayotte 1970	✓	X	✓
Simmons & Shephard 1971	✓	✓	X (invalid correction)
Pugh 1972	✓	✓	X (correction omitted as insignificant)

4. Since lung tissue absorbs some foreign gas, the uptake by pulmonary blood should be calculated from the observed (total) uptake, minus an amount removed by lung tissue.

A review of the literature (Table 6.1) indicated that many studies did not allow for one or more of these precautions.

In the nitrous oxide rebreathing method which is described in this chapter, the above-mentioned precautions were incorporated through the continuous measurement of nitrous oxide and argon gas on a mass spectrometer. Changes in the argon concentration during the rebreathing period indicated the adequacy of gas mixing between the lung and rebreathing bag, and were also used to calculate the total lung-bag volume at appropriate times during the rebreathing manouvre. The simultaneous measurements of the N_2O concentration were then used to calculate cardiac output, as described in Chapter 2 (Section C).

SUBJECTS and METHODS

Nine normal subjects volunteered for the study, and details of their anthropometry are shown in Table 6.2.

Each subject exercised on a bicycle ergometer for periods of half an hour at a fixed work load of 300, 600 or 900 kpm/min (50, 100 and 150 watts). The work loads were generally completed in one day with adequate rest periods between work loads. In some subjects one of the work loads was carried out on a separate day.

Table 6.2: Anthropometric data for the subjects examined in the study.

SUBJECT	AGE (yrs)	SEX	Ht (cm)	Wt (kg)
MM	37	M	171	88.0
RHTE	34	M	173	65.0
GH	33	M	184	85.9
DN	29	M	193	97.0
EZF	24	M	173	70.0
JR	35	M	171	74.9
PH	21	M	170	68.6
PS	24	M	178	72.1
DB	23	M	187	78.0

During each exercise test ECG, ventilation and gas exchange were monitored continuously. A mass spectrometer was used with an on-line computer to facilitate the measurement of all gas concentrations during the rebreathing period. After an initial cycling period of five minutes, a series of ten alternate CO_2 and N_2O rebreathing procedures (each of 10-15 seconds duration) were carried out at intervals of 3 minutes. A continuous record of heart beat and ventilation ensured a return to steady state conditions before the next rebreathing procedure. An off-line print-out of the exercise data subsequently confirmed the return to steady state of various parameters, and also provided the information necessary for the calculation of cardiac output. (Chapter 2: Section C).

Only data which is relevant to the N_2O rebreathing method is presented in this chapter. A comparison between the N_2O and CO_2 rebreathing methods will be presented in Chapter 7.

RESULTS

The oxygen consumptions for a given work load were similar in all subjects (Table A6.1, Appendix). The cardiac outputs ($\dot{Q}N_2O$) for a given oxygen consumption were also similar in all subjects (Figure 6.1) despite differences in their heart rates and stroke volumes (Table A6.1, Figure 6.2).

The intra-subject coefficients of variation of oxygen consumption and heart rate (C.V.) were low at all work loads. The average coefficient of variation and its range in the subjects is given in

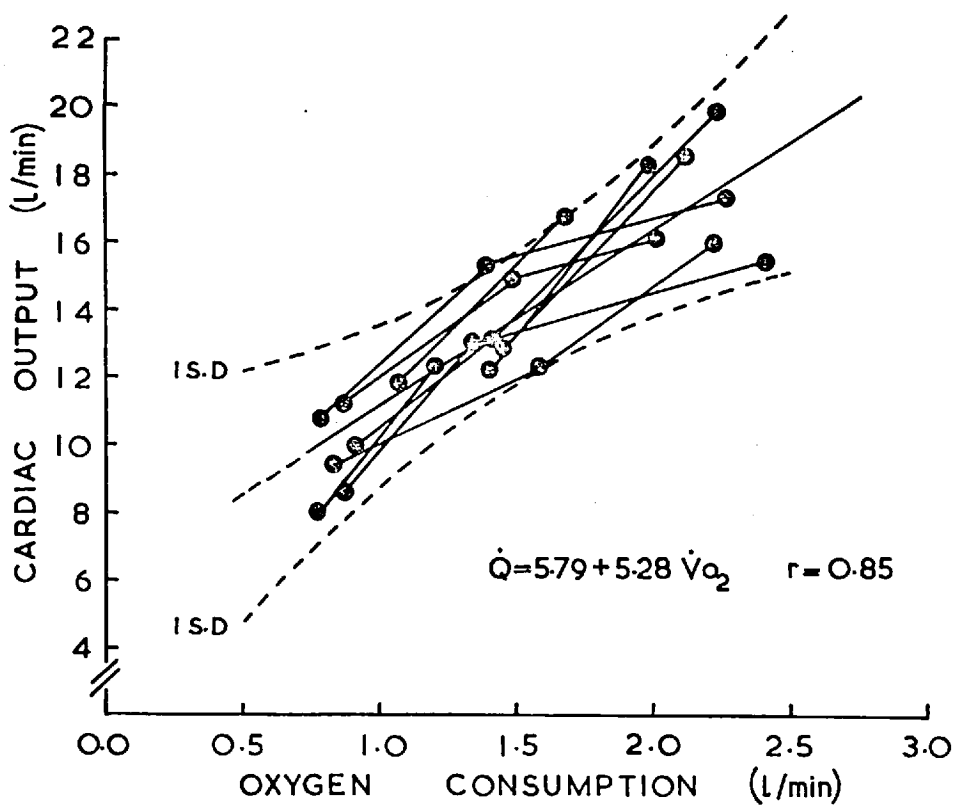


Figure 6.1 Mean values of cardiac output determined by the modified N_2O rebreathing method in seven subjects at each of 3 (or less) work loads, are plotted against the mean values of oxygen consumption in each subject. A regression line (± 1S.D) is also presented for the group data.

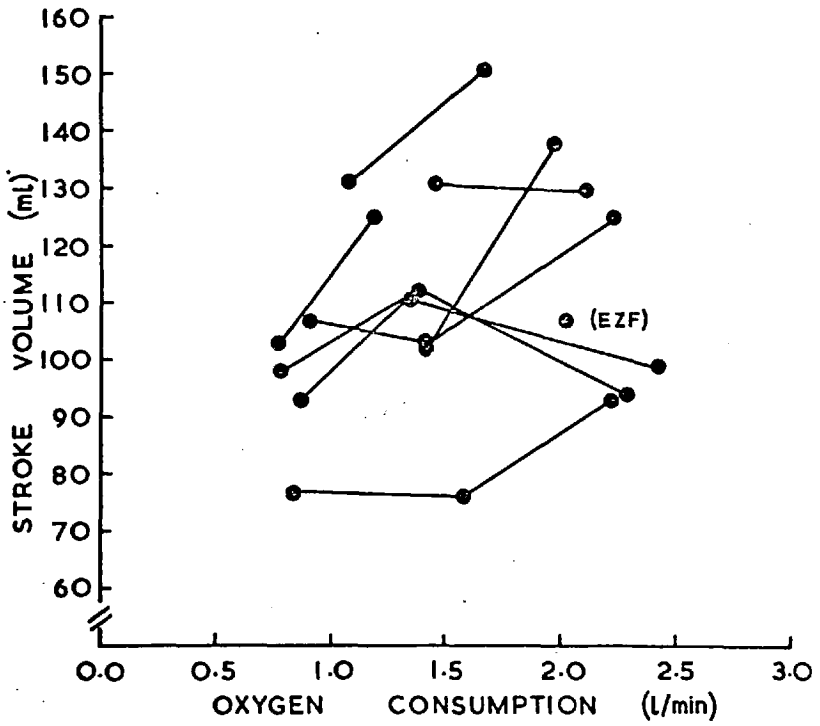


Figure 6.2 Mean values of stroke volume in seven subjects at 3 (or less) work loads are plotted against the mean values of oxygen consumption in each subject.

Table 6.3. The mean intra-subject coefficients of variation of cardiac output and stroke volume (and their range) are also presented. The coefficients of variation of cardiac output were generally lower than 11% at all work loads (Figure 6.3) with the single exception of subject P.S. The mean values at 50, 100 and 150 watts were 7.9%, 6.6% and 5.6% respectively, (corresponding to standard deviations of 0.75, 0.88 and 0.97l/min), while the range of variation about these mean values was reduced with increasing work loads (Figure 6.3). The variations in the cardiac output were also reflected in the coefficients of variation of stroke volume which are shown for each subject in Figure 6.4.

Some measure of the technical variation of the present method was assessed from the coefficient of variation for a paired estimate (C.V.P.E., the adequacy of which has been discussed in Chapter 2, Section d). The C.V.P.E. for cardiac output was 7.0% at a work load of 50 watts, and was reduced to 2.0% and 2.9% at the higher work loads of 100 and 150 watts (Table 6.4).

DISCUSSION

The present method was developed to reduce technical sources of error in the measurement of cardiac output by the N_2O rebreathing method. As the coefficient of variation from paired estimates (C.V.P.E.) has been used widely in the past to express the variability of a measurement, it is used here to compare the present results

SUBJECT	WITHIN SUBJECT COEFFICIENTS of VARIATION (C.V.%)							
	OXYGEN CONSUMPTION (C.V.%)		CARDIAC OUT (C.V.%)		HEART RATE (C.V.%)		STROKE VOLUME (C.V.%)	
	mean	range	mean	range	mean	range	mean	range
MM	3.0	2.5 - 3.5	3.1	2.1 - 4.1	4.1	2.5 - 5.7	3.1	2.6 - 3.5
RHTE	1.3	1.0 - 1.6	7.1	3.9 - 10.3	1.6	1.1 - 2.2	9.7	9.3 - 10.0
GH	3.0	1.3 - 4.6	8.0	3.5 - 10.9	3.0	2.4 - 3.6	7.0	1.3 - 11.0
DN	3.6	1.7 - 5.4	7.8	5.6 - 9.9	7.5	2.3 - 5.2	6.5	4.1 - 8.9
EZF	4.9	1.8 - 6.6	5.0	4.7 - 5.5	-	-	-	-
JR	4.4	2.1 - 8.2	5.8	3.7 - 7.5	3.3	1.1 - 5.0	4.7	3.0 - 7.2
PH	7.1	2.2 - 10.1	5.8	3.8 - 7.2	2.6	1.2 - 3.3	5.0	3.2 - 6.6
PS	5.7	2.6 - 7.4	10.8	6.1 - 16.1	3.5	2.7 - 4.5	11.9	10.6 - 14.3
DB	5.4	4.5 - 6.2	5.6	5.1 - 6.1	3.9	3.2 - 4.6	8.3	9.6 - 9.0

Table 6.3: Showing the intra-subject coefficients of variation of oxygen consumption, cardiac output, heart rate and stroke volume. For each subject, the mean coefficient of variation and its range at three work loads is presented.

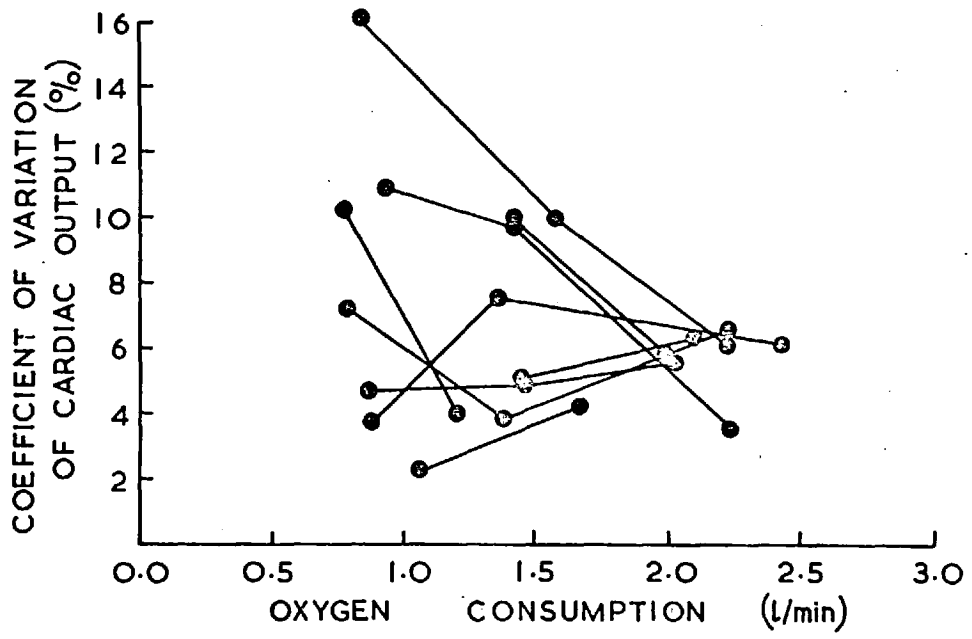


Figure 6.3 The coefficients of variation of cardiac output in seven subjects exercising at several work loads, plotted against corresponding mean values of oxygen consumption.

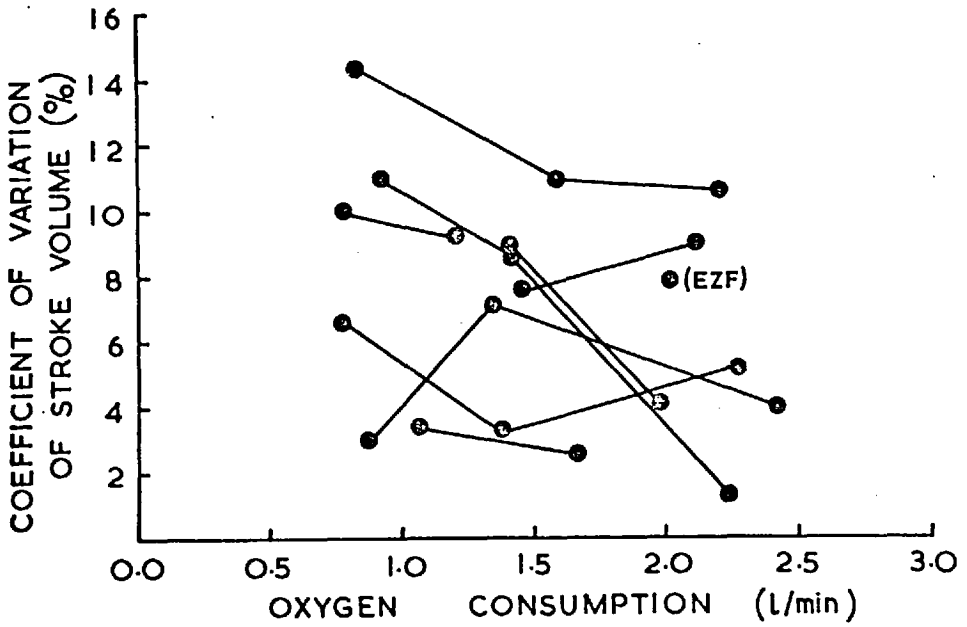


Figure 6.4 The coefficients of variation of stroke volume in seven subjects exercising at several work loads, plotted against corresponding mean values of oxygen consumption.

with previous studies. The 2-7% C.V.P.E. of cardiac output in this study was considerably smaller than that of the modified Cander and Forster (1959) and Ayotte et al. (1970) methods. (6-13%, Chapter 5; Table 6.4). The reduced variability suggests that the technical errors in the measurement of lung-bag rebreathing volume and gas analyses have been considerably reduced.

SOURCES of OVERALL VARIABILITY in the PRESENT METHOD: The coefficient of variation (C.V.) was used to measure the overall biological and technical variation of the present method. At the first work load the high range of variation coefficients for cardiac output (mean : 7.1%, with a range of 2.1-16.1%) was probably due in part to the high variation in subject P.S. However a source of technical error may exist in the use of a large rebreathing bag (4l) at low work loads. It is noted that the accuracy of the method depends on the correct measurement of N_2O uptake from simultaneous changes in the total rebreathing volume, and its N_2O concentration. It is possible to alter the initial bag volume in relation to the subject's tidal volume so that mixing is improved while changes in rebreathing volume and N_2O concentration are optimised.

Further variations in the estimate of cardiac output may have occured because the estimates were not standardised with respect to oxygen consumption. Such a procedure would have required the interpolation of cardiac output measurements for specific values of oxygen consumption and would have been statistically incorrect for

the present data. A small part of the variation in cardiac output was thus due to concomitant changes in the oxygen consumption at the time of measurement.

OTHER SOURCES OF VARIATION: The assumption of a fixed value for the solubility constant of N_2O in blood (0.474 ml/ml/Atmosphere at 37°C) may have caused further errors in measuring cardiac output. An increase in deep body temperature from 37°C (at rest) to about 39°C during heavy exercise (Nielsen, 1938) should raise the value of the solubility constant, so that the assumption of a constant temperature would over-estimate cardiac output by 0.6 l/min at high work loads, while the reproducibility of the method would remain unaffected.

COMPARISON OF VARIABILITY WITH THAT OF OTHER METHODS: The mean intra-subject coefficient of variation of cardiac output (C.V.) in the present method was 5-8% at all work loads. This was considerably smaller than the 7-13% variation (C.V.) of the Ayotte method (Ayotte et al. 1970; 1973) and was comparable to the 5-6% C.V. of the indirect (CO_2) Fick method, as well as the 3-5% C.V. of the dye dilution and direct (O_2) Fick methods (Grimby, 1966; Ekelund and Holmgren, 1964).

The reproducibility of the method was as good or better than that of other foreign gas rebreathing methods. Becklake et al. (1962) using a steady state N_2O method, found a coefficient of variation of 8.5% (C.V.P.E.) for duplicate estimates of cardiac output. Although cardiac output was measured at several work loads the range of variation at different levels of exercise was not given.

(Table 6.4). The C.V.P.E. indices for cardiac output are somewhat better when the acetylene rebreathing method is used (Saltin 1964a, 1964b; Simmons and Shepard, 1971). The 5.9-6.4% C.V.P.E.s found by Saltin were similar to the values in this study, however they hide a consistent error of 2-9% in the measured cardiac output, since the total uptake of acetylene was not corrected for lung tissue absorption. The study of Simmons and Shepard (1971) quoted a coefficient of variation of 2.0-2.5% for paired estimates of the arterio-venous oxygen difference of blood. The authors subsequently treated this variation as the variation in measured cardiac output. However, this was incorrect since cardiac output is also affected by variations in oxygen consumption (Fick equation), and implies that the C.V.P.E. was larger than the quoted figures. Although the reproducibility of the method remains impressive, it is unfortunate that the authors made an arbitrary 10% correction to the measured arterio-venous oxygen difference in order to account for the effect of lung tissue absorption of N_2O (Table 6.1). Logically the correction should be made on a sliding scale of between 2-9% (Cander and Forster, 1959) because the fixed volume of lung tissue will take a smaller fraction of thetotal gas uptake as cardiac output increases.

COMPARISON OF ABSOLUTE VALUES OF CARDIAC OUTPUT: The cardiac output at a given oxygen consumption was similar in all subjects (Figure 6.1). The equation for the cardiac output regression line was $\dot{Q}N_2O = 5.79 + 5.28\ \dot{V}O_2$ ($r = 0.85$). Although the slopes of cardiac output versus oxygen consumption were slightly flattened at high work loads in three subjects, the effect was not marked. However,

Table 6.4: The coefficient of variation of paired estimates of cardiac output (C.V.P.E.%) is used to assess the variability of the modified N_2O rebreathing method. Variability is compared with that of other non-invasive and invasive methods.

SOURCE	METHOD of MEASURING CARDIAC OUTPUT during exercise	COEFFICIENT of VARIATION of PAIRED ESTIMATES of CARDIAC OUTPUT C.V.P.E.%
PRESENT STUDY	Modified N_2O rebreathing method	2.0 - 7.0%
Zeidifard: Ch.5	N_2O rebreathing method (Cander)	6.5 - 10.2%
	N_2O rebreathing method (Ayotte)	7.2 - 13.2%
Becklake et al. 1962	N_2O method (steady state)	8.5% (range not given)
Saltin, 1964	C_2H_2 rebreathing method	5.9 - 6.4%
Simmons & Shephard 1971	C_2H_2 rebreathing method	>2.0 - 2.5%*
Zeidifard: Ch.4	Indirect (CO_2) Fick method	2.3 - 4.4%
Holmgren & Pernow 1960	Direct (O_2) Fick method	5.2%
Ekblom, 1969	Dye dilution method	3.0 - 3.5%

* The value of C.V.P.E. refers to the arterio-venous oxygen difference during exercise. The value for cardiac output would have been higher due to variation in oxygen consumption.

Table 6.5: Showing values of cardiac output measured by various invasive and non-invasive methods in normal subjects. The values of cardiac output were estimated at oxygen consumptions of 1 and 2 l/min, using given (or derived) regression data from each study.

SOURCE*	No. of observations (n)	SUBJECTS	METHOD	CARDIAC OUTPUT (l/min) at an OXYGEN CONSUMPTION of	
				1 l/min	2 l/min
Present study	111	Adults	$\dot{Q}N_2O$	11.1	16.4
Rigatto et al. (1968)	37	Adults	$\dot{Q}N_2O$	11.0	17.8
Becklake et al. (1965)	?/48	Adults (20-40yrs)	$\dot{Q}N_2O$	10.9	15.0
Seely et al. (1974)	504	Children	$\dot{Q}N_2O$	10.1	16.7 (extrapolated value)
Ayotte et al. (1973)	26	Adults	$\dot{Q}N_2O$	11.3	15.6
Simmons and Shephard (1971)	82	Adults	$\dot{Q}C_2H_2$	10.4	16.0
Saltin (1964a, 1964b)	23	Adults	$\dot{Q}C_2H_2$	10.9	15.3
Collected data (Zeidifard)	209	Adults	DYE	11.1	15.8
Collected data (Zeidifard)	56	Adults	FICK O_2	10.8	16.6
Godfrey et al. (1971)	161	Children	$\dot{Q}CO_2$ uncorrected	9.6	15.2
Present study	102	Adults	$\dot{Q}CO_2$ uncorrected	10.0	14.8

* The sources of collected data are presented in Table A8.1.

it has been observed by previous workers (Saltin, 1964a; Astrand et al. 1964) at work loads greater than 40-70% of a subject's maximum working capacity.

The present values of cardiac output were all measured during exercise, and were compared to corresponding values in the literature (Table 6.5). Where investigators had presented individual values of cardiac output at rest and exercise, the measurements made during exercise were collected to recalculate the regression of cardiac output against oxygen consumption for exercise alone. The cardiac outputs measured in this study compared favourably with values found by other workers using the foreign gas and CO_2 rebreathing methods. They were also very similar to those based on the collected data (Zeidifard) from dye dilution and direct (O_2) Fick methods. The present N_2O method is therefore seen to give estimates of cardiac which are both reliable and generally reproducible. However, since there are several foreign gas methods currently in use, it is obviously desirable to know the advantages or disadvantages of these methods.

CRITICISM OF METHODS: The limitations of the nitrous oxide rebreathing methods of Ayotte et al. (1970) and Cander and Forster (1959) were examined in Chapter 5 and are not included in the present discussion. The choice then remains of using the present nitrous oxide rebreathing method, an acetylene rebreathing method, or the open circuit N_2O method of Becklake et al. (1962). In making a choice, the same factors which prompted the modifications in the present method need to be satisfied, namely:

1. Is it better to use acetylene or nitrous oxide to measure pulmonary blood flow?

2. Is the source of foreign gas fully mixed in the lung prior to the measurement of gas uptake?

3. How effectively are changes in lung-bag volume and/or gas uptake measured?

4. Is recirculation time satisfactorily recorded and allowed for?

The concentrations of N_2O which are used in the above methods are fairly harmless since the rebreathed concentrations (approx. 5%) and the period of exposure are both small. N_2O gas is also reasonably safe to handle and analyse with fast-response electronic equipment. Two advantages which derive from this are that a) nitrous oxide concentrations can be monitored continuously throughout a given manouvre, and b) repetition of the manouvre is not limited by analytical or technical methodology.

In contrast the use of acetylene is disadvantageous in several respects. Acetylene gas is explosive in the presence of oxygen, and is also unpleasant to inhale. Because of the safety considerations, acetylene analysers which can continually monitor concentrations greater than 1-2% are slow response instruments. A fast-response instrument such as a mass spectrometer is unsafe since its sampling system operates at high temperatures. The objections to acetylene are overcome if dilute concentrations (1-2%) of the gas are used.

However, although such concentrations are not unpleasant to the subject, they can only be analysed by chemical or chromatographic techniques which are laborious and time-consuming. This immediately places the acetylene rebreathing method at a disadvantage, since only contiguous samples (taken at specific times) can be made. During the rebreathing manouvre an early termination of the mixing period (within 4-5 seconds) is not guaranteed experimentally, and can only be assumed with the patients' co-operation. In addition the subsequent changes in the lung-bag volume during the rebreathing period are determined from two single samples of bag gas taken at 4-5 seconds (after mixing) and at 10 seconds, prior to recirculation. Coupled with the long delay in the estimation of acetylene, the procedure becomes onerous for routine use.

By using an open circuit N_2O breathing method, Becklake et al. (1962) have been able to use fast-response instruments for a continuous record of N_2O during the breathing manouvre. However, their method does raise several objections. During a 20 second period when 5-7% N_2O is breathed, their subjects breathe at a fixed frequency, subsequent to an initial period of rapid breathing to mix inspired and alveolar gases. Nevertheless, the assessment of mixing is not accurate, and is gauged from a steady end-tidal N_2O trace. The effect of recirculation is also not directly detectable, and is noted from a gradual decline in the estimated cardiac output. The uptake of N_2O by pulmonary blood is itself incorrectly calculated, since the total uptake (used to measure $\dot{Q}$) is not corrected for N_2O absorbed by lung tissue. In addition, this uptake is not corrected

for changes in the respiratory exchange ratio (R) since R is assumed to be 1 on the grounds that "fluctuations in end-expiratory levels usually excess the smaller differences between O_2 uptake and CO_2 output". However in the presence of nitrous oxide R is appreciably different from 1 due to the cumulative uptake of N_2O and O_2 in exchange for CO_2 (output).

In contrast, the present N_2O rebreathing method has attempted to eliminate any assumption which could contribute to the variability of the method in routine use. The use of a mass spectrometer to continually measure all gases during a rebreathing period, allows a frequent repetition of the method. The simultaneous measurements of argon and N_2O ensures that the total uptake of N_2O is calculated after the initial mixing period, and is corrected for changes in the rebreathing volume (or R). Prior to the estimation of cardiac output, the N_2O absorbed by lung tissue is subtracted from the total uptake of the gas, while significant recirculation of pulmonary blood is readily detectable from the trace of the alveolar N_2O concentration.

CONCLUSIONS

The present N_2O rebreathing method was developed in order to measure cardiac output reliably, while reducing the errors and variability of the foreign gas methods as used by previous workers. During exercise, the method was found to give values of cardiac output which were comparable to, and as reproducible as values measured by the direct (O_2) Fick and dye dilution methods.

CHAPTER 7

A COMPARISON OF THE PLATEAU CO_2 (INDIRECT FICK) AND NITROUS OXIDE REBREATHING METHODS FOR MEASURING CARDIAC OUTPUT

INTRODUCTION

Previous chapters in this thesis have examined several carbon dioxide and nitrous oxide rebreathing methods for the measurement of cardiac output.

The plateau CO_2 (indirect Fick) rebreathing method and the modified N_2O rebreathing method were found to give reliable and reproducible estimates of cardiac output, which compared favourably with values measured by the direct (O_2) Fick and dye-dilution methods (Chapters 4 and 6).

In this chapter these two rebreathing methods were compared with each other when used alternately on normal subjects during sustained periods of exercise on a bicycle ergometer.

SUBJECTS AND METHODS

The experimental design of the study, and the subjects involved have been discussed in Chapter 6.

Nine subjects were exercised on a bicycle ergometer for periods of half an hour at each of three work loads (50, 100 and 150 watts). At each work load the plateau CO_2 and N_2O rebreathing methods were used alternately (at intervals of 3 minutes) for a total of ten measurements of cardiac output. The procedures for the calculation of cardiac output by the two methods have been described in sections (a) and (c) of Chapter 2. When the CO_2 rebreathing method was used, cardiac output was calculated both

before and after a nominal correction was applied to the measured plateau $F\bar{V}CO_2$, due to the "downstream effect" discussed in Chapters 1 and 2.

RESULTS

The values of cardiac output based on the nitrous oxide rebreathing method have been presented in table A6.1 of the Appendix. Corresponding values of cardiac output from the CO_2 rebreathing method are shown in Appendix table A7.1. The table presents repeated measurements of oxygen consumption in individual subjects, during a steady state of exercise at each of three work loads. Each measurement was made before a CO_2 rebreathing procedure.

Repeated estimates of cardiac output ($\dot{Q}CO_2$) in each subject are shown with the corresponding oxygen consumption. In each instance two values of cardiac output are given, based on "downstream" corrected and uncorrected values of plateau $F\bar{V}CO_2$.

The mean values of oxygen consumption and cardiac output (together with their coefficients of variation) are given for individual subjects at each work load. Due to technical faults in the apparatus, no data are available at some work loads in subjects MM, RE, DN and DB.

The reproducibility of the N_2O and CO_2 rebreathing methods of measuring cardiac output was assessed in table 7.1. With the N_2O rebreathing method the coefficients of variation of paired estimates of cardiac output (C.V.P.E.) were in a range of 2-7% at all levels of exercise. The mean overall variation within subjects (C.V.) was only slightly higher, at 5.6 - 7.9%.

Table 7.1: Shows the variability of cardiac output measured in nine subjects, using the modified N_2O and plateau CO_2 rebreathing methods during exercise. Variability is measured by the C.V.P.E. and C.V. indices, and in each case the range of variation from several work loads is presented.

METHOD of MEASURING CARDIAC OUTPUT	COEFFICIENT of VARIATION of PAIRED ESTIMATES of CARDIAC OUTPUT (C.V.P.E.%)	Range of the mean COEFFICIENT of VARIATION of CARDIAC OUTPUT WITHIN SUBJECTS (C.V.%)
Present study: Modified N_2O rebreathing method	2.0 - 7.0 %	5.6 - 7.9 %
CO_2 rebreathing method	1.6 - 3.6 %	6.9 - 8.8 %

The variation coefficients for cardiac output measured by the CO_2 rebreathing method were similar, with a C.V.P.E. of 1.6 - 3.6%, and a mean intrasubject C.V. range of 6.9 - 8.8% at all levels of work (Table 7.1). The downstream correction for $\bar{P}vCO_2$ had only a small effect on the variability of cardiac output estimates (Table A7.1), so that the coefficients of variation (C.V.) presented in table 7.1 are representative of both corrected and uncorrected $\dot{Q}CO_2$.

For each subject the mean values of $\dot{Q}N_2O$, $\dot{Q}CO_2$ uncorr. and $\dot{Q}CO_2$ corr. were plotted separately against corresponding mean values of oxygen consumption at each work load (Figure 7.1, 7.2 and 7.3). The regression lines for the group are also presented, and are plotted separately in Figure 7.4 to facilitate comparison.

The regression of $\dot{Q}N_2O$ against oxygen consumption ($\dot{Q}N_2O$ = 5.79 + 5.28 $\dot{V}O_2$) compared favourably with that of $\dot{Q}CO_2$ uncorr. ($\dot{Q}CO_2$ uncorr. = 5.16 + 4.81 $\dot{V}O_2$); however, $\dot{Q}N_2O$ values were generally 1.0 - 1.6 l/min higher at measured levels of oxygen consumption. In comparison the $\dot{Q}CO_2$ corr. regression line ($\dot{Q}CO_2$ corr. = 6.30 + 6.01 $\dot{V}O_2$) was steeper and gave values of cardiac output which were 1.0 - 2.2 l/min higher than corresponding $\dot{Q}N_2O$ values.

The alternated estimates of $\dot{Q}N_2O$ and $\dot{Q}CO_2$ in each subject were plotted in Figures 7.5 and 7.6 (using $\dot{Q}CO_2$ uncorr. and $\dot{Q}CO_2$ corr. values respectively). In each case regression lines for the data are compared against a line of identity. The regression line of $\dot{Q}N_2O$ against $\dot{Q}CO_2$ uncorr. lay close to the line of identity ($\dot{Q}N_2O$ = 2.69 + 0.90 $\dot{Q}CO_2$), whereas that of $\dot{Q}N_2O$ against $\dot{Q}CO_2$ corr. differed appreciably ($\dot{Q}N_2O$ = 3.47 + 0.67 $\dot{Q}CO_2$).

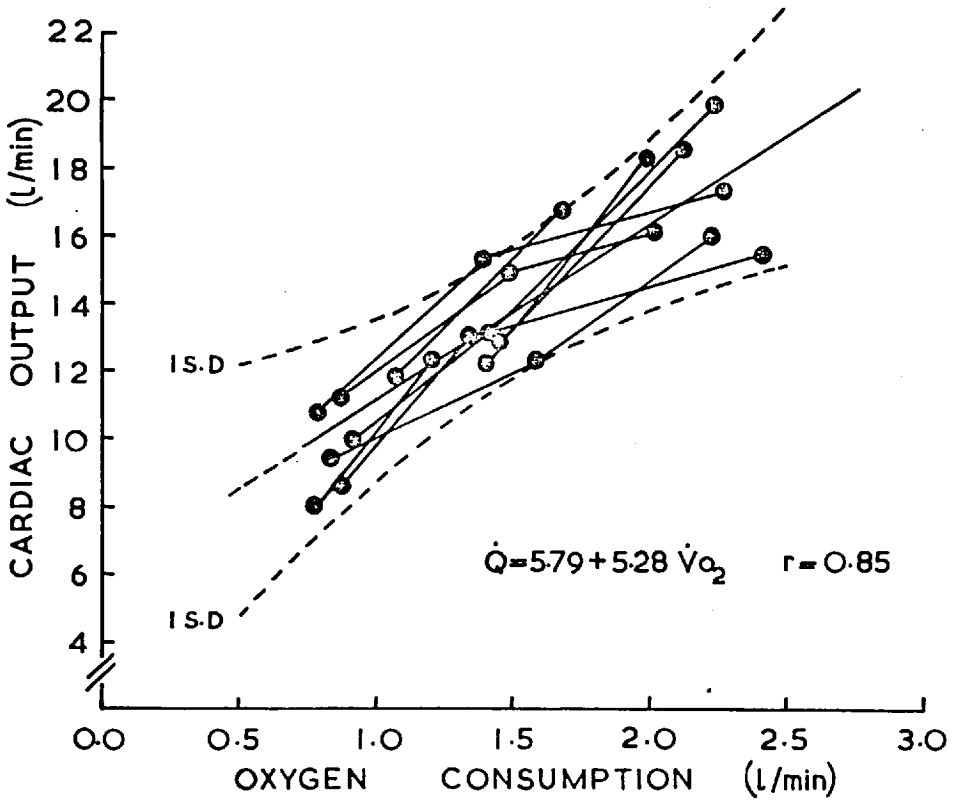


Figure 7.1 Mean values of cardiac output determined by the modified N_2O rebreathing method at each of 3 (or less) work loads in seven subjects, plotted against the mean values of oxygen consumption in each subject. A regression line (± 1S.D) is presented for the group data.

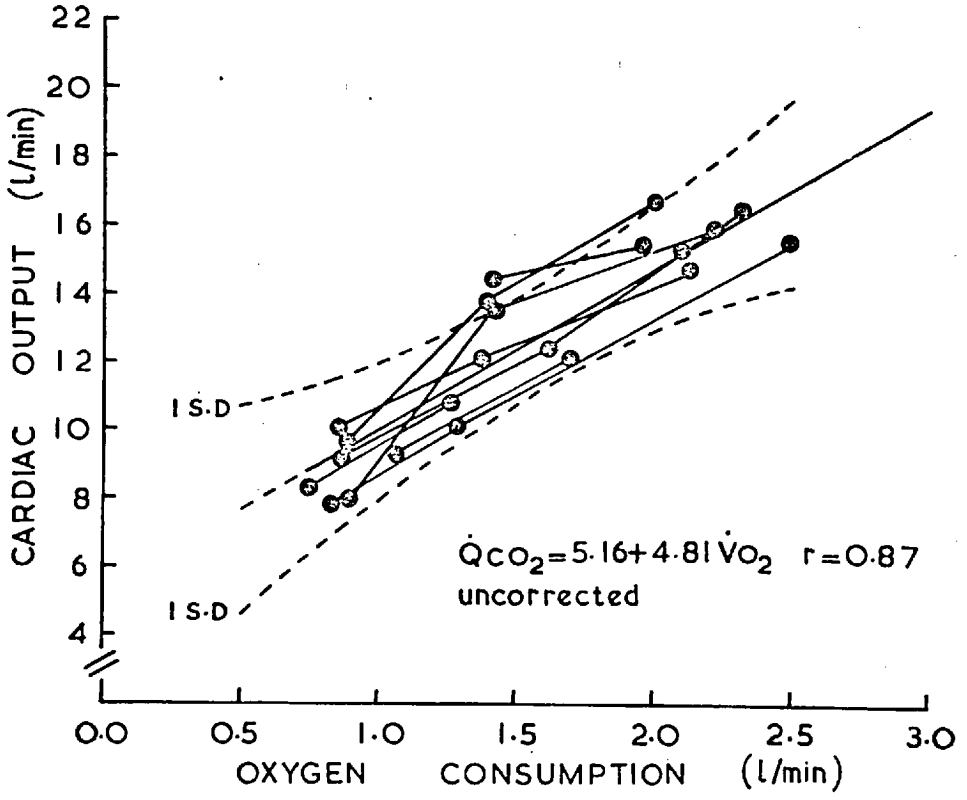


Figure 7.2 Mean values of cardiac output determined by the indirect (CO_2) Fick method in seven exercising subjects (using plateau $P\bar{v}CO_2$ values uncorrected for the "downstream" effect), are plotted against the mean values of oxygen consumption at each work load. A regression line ($\pm$1S.D) for the group is also presented.

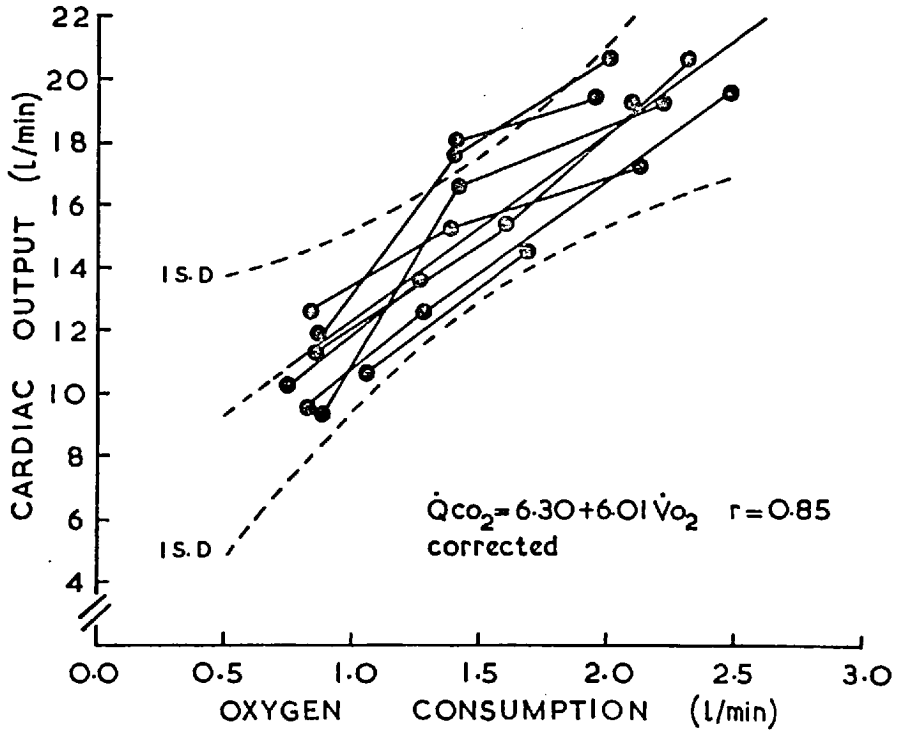


Figure 7.3 Mean values of cardiac output determined by the indirect (CO_2) Fick method in seven exercising subjects (using values of plateau $P\bar{v}CO_2$ corrected for the "downstream" effect), are plotted against the mean values of oxygen consumption at each work load. A regression line ($\pm$ 1S.D) is presented for the group data.

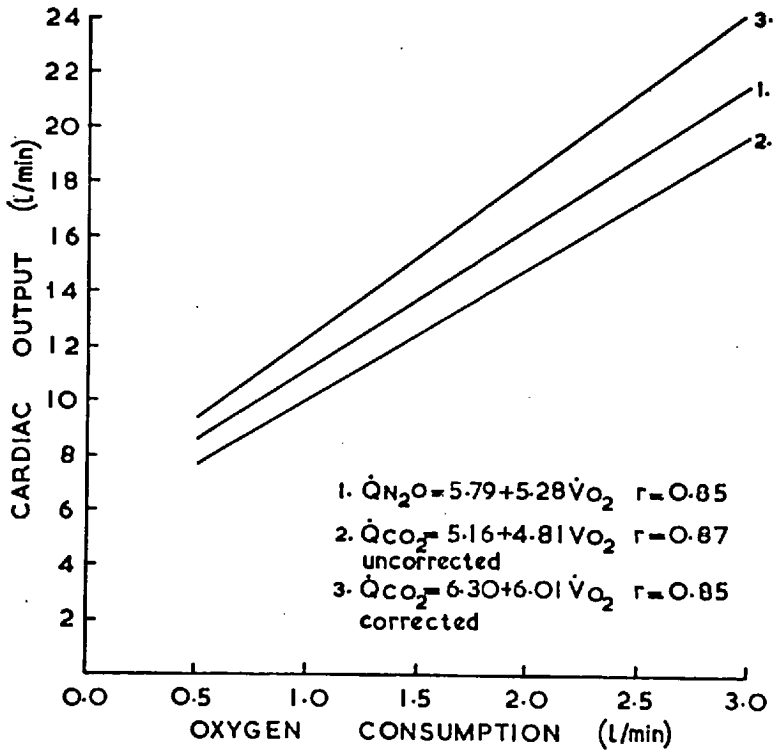


Figure 7.4 Regression lines for the group data of cardiac output plotted against oxygen consumption. Cardiac output was determined by (1) the modified N_2O rebreathing method (2) the indirect (CO_2) Fick method using values of $P\bar{v}CO_2$ uncorrected for the "downstream" effect (3) the indirect (CO_2) Fick method using values of $P\bar{v}CO_2$ corrected for the "downstream" effect.

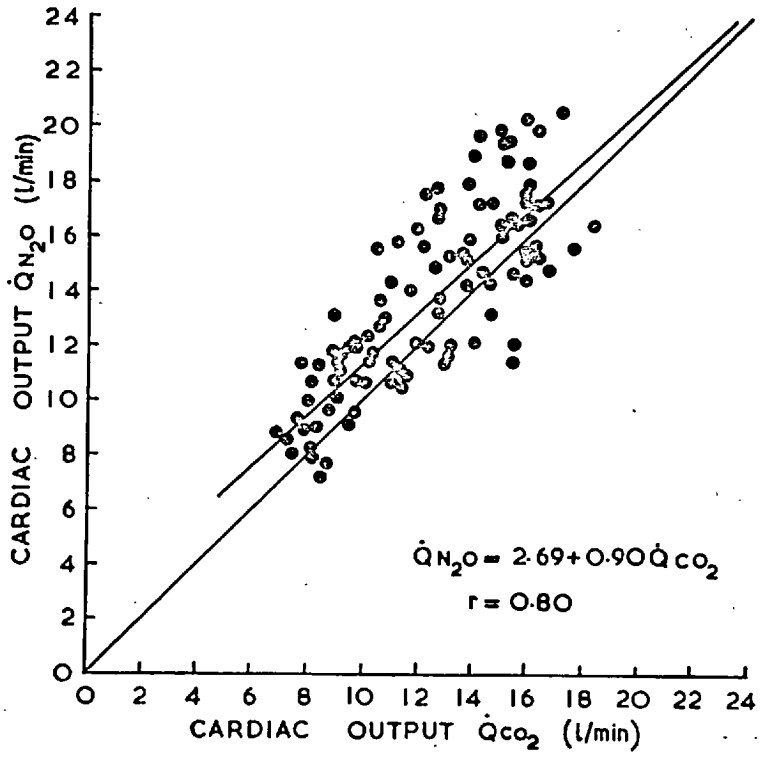


Figure 7.5 Concordance plot for paired estimates of cardiac output determined by the $\dot{Q}N_2O$ and $\dot{Q}CO_2$ ("uncorrected") rebreathing methods within 3 minutes of each other. A regression line for the data is also presented.

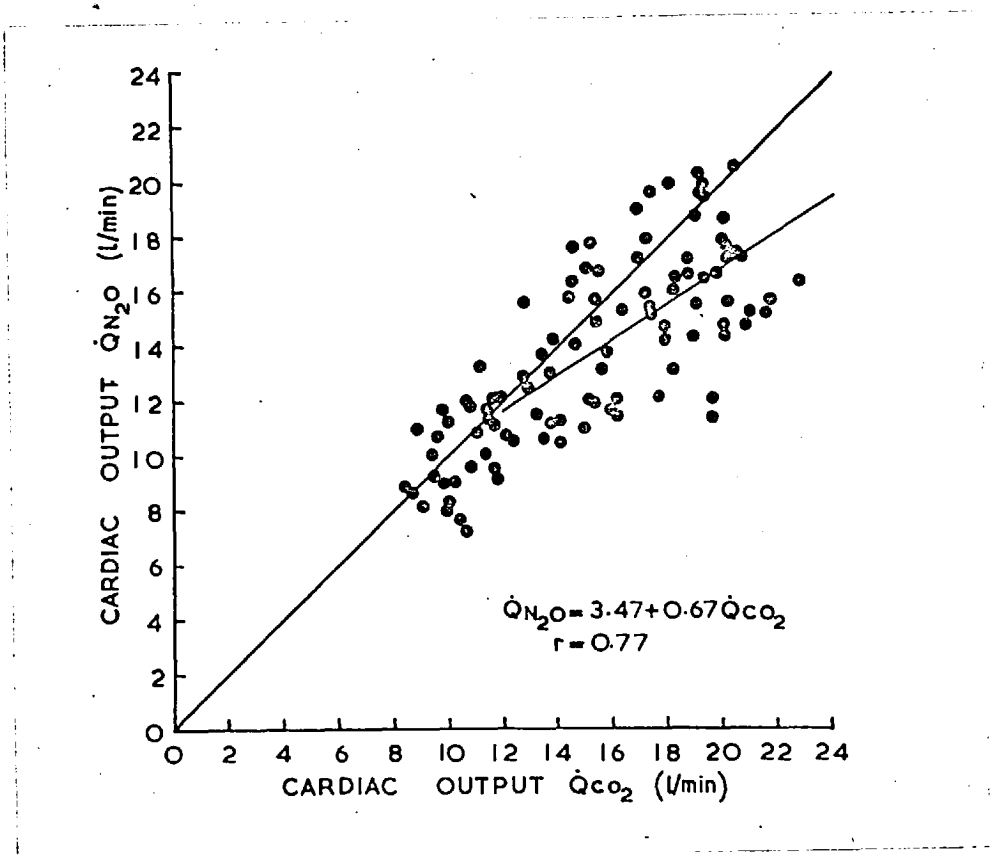


Figure 7.6 Concordance plot for paired estimates of cardiac output determined by the $\dot{Q}N_2O$ and $\dot{Q}CO_2$ ("Corrected") rebreathing methods within 3 minutes of each other. A regression line for the data is also presented.

DISCUSSION

The variations in cardiac output measured by the N_2O and CO_2 rebreathing method were similar (Table 7.1), and compare favourably with the variability of invasive methods (as discussed in Chapter 4 and 6).

Provided $\dot{Q}CO_2$ values were based on the plateau $\bar{P}vCO_2$ uncorrected for the "downstream effect", the regression lines for cardiac output measured by the two methods were fairly similar, with slight differences in slope and intercept (Figure 7.4). A "downstream" correction to $\bar{P}vCO_2$ raised the values of $\dot{Q}CC_2$, so that both the slope and intercept of the $\dot{Q}CO_2$ corr. regression line were raised above those of the $\dot{Q}N_2O$ line.

The use of a "downstream" corrected value of $\dot{Q}CO_2$ is contra-indicated for two reasons. Firstly, it was noted in chapter 4 that no great significance was attached to the slightly low values of $\dot{Q}CO_2$ uncorr. As before, $\dot{Q}CO_2$ in the present study was calculated from a value of $PaCO_2$ based on (and related to) the expected normal physiological dead space of each subject. The latter was derived from the comprehensive study of Godfrey et al. (1971), which indicated that resting dead space increased by 5 ml for every 100 ml increase in tidal volume during exercise. However, if an increase of 8-11 ml/100 ml was assumed from the limited studies of Asmussen and Nielsen (1952), Jones et al. (1966) and Taylor (1972), $PaCO_2$ would have been raised, with a corresponding increase of 0.4 - 0.8 l/min in cardiac output at different work loads. This increase would have made the regression lines of $\dot{Q}N_2O$ and $\dot{Q}CO_2$ uncorr. almost identical, while that of $\dot{Q}CO_2$ corr. would have been raised even further. The second objection to the use of "downstream" corrected values of cardiac output is the body of

evidence in the literature; the values of $\dot{Q}CO_2$ corr. are too high when compared with values measured by both invasive and non-invasive methods, as indicated in Table 7.2.

It seems therefore that both the N_2O and CO_2 rebreathing methods (using uncorrected values of $\bar{P}vCO_2$) are equally reproducible, and are equally reliable in measuring cardiac output.

The two rebreathing methods were further examined in concordance plots of the almost simultaneous measurements of $\dot{Q}N_2O$ and $\dot{Q}CO_2$ (both uncorrected and corrected) shown in Figures 7.5 and 7.6. The position of a point on such a plot would depend on the total variability of paired estimates of cardiac output, and includes both the technical variability of the method, and the biological variation of the subjects.

In the comparison of $\dot{Q}N_2O$ with both $\dot{Q}CO_2$ uncorr. and $\dot{Q}CO_2$ corr., 68 out of 101 observations were within $\pm 20\%$ limits of the line of identity. However, if values of $\dot{Q}CO_2$ were raised by about 1 litre to simulate the effect of a slightly enlarged dead space (as discussed earlier), then 75/101 paired estimates of $\dot{Q}N_2O$ and QCC_2 uncorr. would fall within the $\pm 20\%$ line of identity; the corresponding value for $\dot{Q}CO_2$ corr. would be 53 out of 101 paired estimates.

The concordance plots in Figures 7.5 and 7.6 differ from similar data published in the literature (Table 7.3). With the exception of Klausen (1965), no studies have compared the simultaneous (or almost simultaneous) use of two non-invasive procedures for measuring cardiac output. In most cases the non-invasive technique was compared against the dye dilution method.

Table 7.2: Showing values of cardiac output measured by various invasive and non-invasive methods in normal subjects. The values of cardiac output were estimated at oxygen consumptions of 1 and 2 l/min, using given (or derived) regression data from each study.

SOURCE*	No. of observations	SUBJECTS	METHOD	CARDIAC OUTPUT (l/min) at an OXYGEN CONSUMPTION of 1 l/min	2 l/min
Collected data (Zeidifard)	209	Adults	DYE	11.1	15.8
Collected data (Zeidifard)	56	Adults	FICK O_2	10.8	16.6
Present data	111	Adults	$\dot{Q}N_2O$	11.1	16.4
Becklake et al. (1965)	?/48	Adults	$\dot{Q}N_2O$	10.9	15.0
Present data	102	Adults	$\dot{Q}CO_2$ uncorrected	10.0	14.8
Godfrey et al. (1971)	161	Children	$\dot{Q}CO_2$ uncorrected	9.6	15.2
Present data	102	Adults	$\dot{Q}CO_2$ corrected	12.3	18.3
Edwards et al. (1969)	12	Adults	$\dot{Q}CO_2$ corrected	12.8	19.4
Denison et al. (1971)	60	Adults	$\dot{Q}CO_2$ corrected	14.9	22.6
Spiro et al. (unpublished)	30	Adults	$\dot{Q}CO_2$ corrected	11.5	17.7

* Sources of collected data are shown in Table A8.1.

Table 7.3: Shows the results of concordance plots for simultaneous (or nearly simultaneous) determinations of cardiac output measured by two independent methods. The sources of information are presented.

STUDY	METHODS COMPARED	No. of subjects	LIMITS of the LINE of IDENTITY	No. of observations	No. of observations within the Line of IDENTITY
Present study	1) $\dot{Q}N_2O$ versus $\dot{Q}CO_2$ uncorr.	9	± 20 %	101	68 (75) *
	2) $\dot{Q}N_2O$ v. $\dot{Q}CO_2$ corr.	9	± 20 %	101	68 (53) *
Klausen, 1965	$\dot{Q}C_2H_2$ v. $\dot{Q}CO_2$ (exponential)	1	± 15 %	67	65
Ayotte et al. 1970	$\dot{Q}N_2O$ v. $\dot{Q}$ dye	4	± 15 %	36	34
Becklake et al., 1962	$\dot{Q}N_2O$ v. $\dot{Q}$ dye	8	± 20 %	26	25

* See discussion.

Table 7.3: continued.

STUDY	METHODS COMPARED	No. of subjects	LIMITS of the LINE of IDENTITY	No. of observations	No. of observations within the LINE of IDENTITY
Asmussen & Nielsen, 1952	$\dot{Q}C_2H_2$ v. $\dot{Q}$ dye	26	$\pm$ 20 %	100	78
Saltin, 1964	$\dot{Q}C_2H_2$ v. $\dot{Q}$ dye	4	$\pm$ 20 %	42	39

In comparison with the present work, a larger proportion of the paired estimates of cardiac output in the above studies lie within ±15 - 20% of the line of identity (Table 7.3). Two factors may have contributed to this; in all the studies (Klausen, 1965 excepted) the determinations of cardiac output by each method were made within seconds of each other. The paired determinations in this study were carried out at intervals of three minutes, thus allowing time for possible biological variations in oxygen consumption and cardiac output. On the other hand Klausen (1965) allowed an interval of 5 minutes between paired determinations of cardiac output, despite which all his estimates were within a ±15% line of identity; however, only one subject was involved in the study, so that the degree of variability was likely to remain within narrow limits.

Noting the limitations of the present study, the concordance of the paired estimates of cardiac output was quite favourable, being similar to the concordance of the acetylene with the dye dilution method (Asmussen and Nielsen, 1952; Saltin, 1964a and b). Nevertheless, the whole procedure of concordance plots becomes somewhat doubtful if it is recalled that the methods of Kalusen (1965), Ayotte et al. (1970), and Becklake et al. (1962) are relatively poor in technical reproducibility (Chapters 3, 4 and 6); in spite of this, comparisons with the dye dilution method are often as favourable (within ±15% of the line of identity) as those based on more reproducible methods examined in this and other studies (Chapter 6; Saltin, 1964a and b; Asmussen and Nielsen, 1952). The explanation is not clear, since almost identical procedures were used

for the paired determinations of cardiac output in all the studies. The number of subjects examined is unlikely to have affected the results since, apart from limiting the range of intra-subject variation, there is no evidence to suggest that there is a subject-specific relationship in the paired estimates of cardiac output which would give a biased advantage to smaller numbers of subjects. On the other hand a greater number of observations would give a more accurate delineation of the relationship of one value of cardiac output with the other. In this context a regression line with a correlation coefficient for the data would have been useful in all of these studies, as the true relationship of the paired values and its variance would have been more accurately defined.

CONCLUSIONS

The present study has shown that almost simultaneous measurements of cardiac output can be determined with comparable reproducibility by both the nitrous oxide and plateau carbon dioxide rebreathing methods. Provided the measured $F\bar{V}CO_2$ is not corrected for the "downstream effect" the values of cardiac output measured by both methods are similar to each other and to the published data from several studies using the direct (O_2) Fick and dye dilution methods during exercise.

CHAPTER 8

GENERAL SUMMARY AND CONCLUSIONS

This thesis has examined several CO_2 and N_2O rebreathing techniques for measuring cardiac output. The reproducibility and reliability of these measurements was assessed in normal subjects during steady state exercise on a bicycle ergometer.

The CO_2 rebreathing method: The CO_2 rebreathing method was used to determine the mixed venous PCO_2 in normal subjects; this value was then inserted into the (CO_2) Fick equation to determine cardiac output measured as total systemic blood flow.

In Chapter 3 the measurement of $P\bar{v}CO_2$ by two basically dissimilar CO_2 rebreathing procedures was evaluated. Exponential rebreathing estimates of $P\bar{v}CO_2$ were compared against plateau, or linearly extrapolated values of $P\bar{v}CO_2$ (in the absence of a true plateau). The reproducibility of the measurements was assessed in normal subjects who were given a range of CO_2 rebreathing mixtures.

It was found that the mean values and variability of linearly extrapolated (and plateau) $P\bar{v}CO_2$ in the group did not alter markedly with changes in the initial CO_2 content of the rebreathing bag. In contrast, exponentially extrapolated values of $P\bar{v}CO_2$ were affected by several factors. Procedures which standardised the exponential rise of PCO_2 against rebreathing time (prior to estimating $P\bar{v}CO_2$) were more reproducible than those which did not. The variability of $P\bar{v}CO_2$ in the group was also affected by the initial CO_2 content of the rebreathing mixture; when the

CO_2 content of the mixture was low (as has been recommended in the literature) the variability of the $\bar{P}vCO_2$ estimates was high compared to plateau values. On the other hand, when the CO_2 concentrations were raised to levels suited to a plateau $\bar{P}vCO_2$ manouvre, the estimates of $\bar{P}vCO_2$ were not necessarily more reproducible than plateau $\bar{P}vCO_2$ values.

It was therefore concluded that $\bar{P}vCO_2$ should be measured by the plateau CO_2 rebreathing method, and by a linear extrapolation procedure in the absence of a plateau. In the last resort, the use of the exponential extrapolation procedure required greater control of both the rebreathing mixture, and the pattern of rebreathing.

In Chapter 4 an assessment was made of the day to day intra-subject variation in plateau $\bar{P}vCO_2$, and of the cardiac output determined from it (using the indirect (CO_2) Fick equation). In common with other invasive and non-invasive methods, the variability of resting estimates of cardiac output was high. This was attributed to the absence of real steady state conditions at rest, and to the small veno-arterial CO_2 content difference of blood in resting subjects.

During exercise the day to day intra-subject variation in $\bar{P}vCO_2$ was considerably reduced (mean coefficient of variation of 2-3%), and was only 1-2% higher than the variation measured in five subjects over a half-hour period (Godfrey and Wolf, 1972). Corresponding values of cardiac output based on plateau $\bar{P}vCO_2$, and their reproducibility (mean intra-subject coefficient of variation of 5-6%) were comparable

to values found by the direct (O_2) Fick and dye dilution methods.

In these experiments cardiac output was calculated both with and without a "downstream" correction to the plateau alveolar $P\bar{v}CO_2$ (Chapters 4 and 7). Absolute values of cardiac output based on "uncorrected" $P\bar{v}CO_2$ were found to be comparable to those quoted in the literature for invasive techniques; on the other hand, a "downstream" correction gave values of cardiac output which were high. The evidence from the present work and from various sources in the literature suggests that these values were unrealistically high, and that the downstream correction to plateau $P\bar{v}CO_2$ was unnecessary.

The N_2O rebreathing method: The N_2O rebreathing method was used to determine cardiac output (measured as pulmonary blood flow) from the uptake of N_2O during a ten-second rebreathing period.

An analysis of two recently proposed N_2O rebreathing methods (the method of Ayotte et al. 1970; and a rebreathing form of the Cander and Forster method, 1960) indicated that they were poor in reproducibility, and under-estimated cardiac output (Chapter 5). The poor results were attributed to several causes, the most important of which was omission of a correction for changes in lung-bag volume during the rebreathing period. In the Ayotte method, errors in the measurement of mean alveolar N_2O concentration by planimetry caused further under-estimation of cardiac output.

Several factors were later found to be important in the correct determination of cardiac output by an N_2O rebreathing method (Chapter 6). Complete mixing of gases in the lungs and rebreathing bag was required before the N_2O uptake (and simultaneous lung-bag volume changes) was measured.

To determine the N_2O absorbed by pulmonary blood, it was also necessary to assess the total N_2O uptake prior to recirculation of blood, and to correct it for a quantity of N_2O absorbed by lung tissue.

An investigation of the literature indicated that most studies had ignored one or more of these conditions. A modified N_2O rebreathing method was therefore developed (Chapter 2c) to permit a more accurate estimation of cardiac output. Using the modified method, the absolute values and variability of cardiac output were found to be comparable to measurements made by the direct (O_2) Fick and dye dilution methods.

Comparison of the CO_2 and N_2O rebreathing methods: The CO_2 and N_2O rebreathing methods were alternated repeatedly to determine cardiac output at 3 minute intervals over a half hour period of exercise (Chapter 7). The reproducibility of the two methods was found to be fairly similar; however the values of cardiac output based on the CO_2 rebreathing method, using uncorrected values of $P\bar{v}CO_2$ ($\dot{Q}CO_2$ uncorrected) were often slightly lower than those based on the N_2O rebreathing method ($\dot{Q}N_2O$). This was not considered significant in view of certain assumptions which were made in deriving arterial PCO_2 to determine $\dot{Q}CO_2$ (Chapters 2a, 4 and 7). Taking account of this difference, a concordance plot of $\dot{Q}N_2O$ against $\dot{Q}CO_2$ uncorrected indicated that paired values of cardiac output were usually within a $\pm 20\%$ line of identity. This should be expected in normal subjects since the values of cardiac output measured as systemic and pulmonary blood flow must be similar.

Possible roles and applications of the CO_2 and N_2O rebreathing methods: Both the CO_2 and N_2O rebreathing methods use non-invasive techniques for measuring cardiac output. Since catheterization is unnecessary, the

two methods can be used as simple research tools, without raising serious ethical objections. They may also have an application in the routine exercise testing of pediatric and adult patients with lung or heart disease (Godfrey, 1970; Taylor, 1972).

In patients with lung disease the plateau CO_2 rebreathing method is the one of choice, since the measurement of $P\bar{v}CO_2$ (and cardiac output derived from it) does not depend critically on the gas mixing pattern of CO_2 (unlike the exponential rebreathing methods). The plateau $P\bar{v}CO_2$ is measured during a period of equilibrium in the CO_2 gas tension of the rebreathing bag, the lungs, and mixed venous blood (prior to recirculation); provided equilibrium is achieved, the condition of the lungs is of secondary importance. In comparison, the N_2O rebreathing method cannot be used effectively in such patients, since adequate mixing is essential to its accuracy.

Theoretically, in patients who have heart disease with minimal lung involvement (or who are post-operative subjects) the two methods could be used almost simultaneously to measure systemic and pulmonary blood flows. In such a procedure the N_2O method would detect the existance of shunts, the sense of which (L to R, or R to L) would be determined from the measured systemic blood flow, and from the patients' previous history. Although the CO_2 rebreathing method has previously been used in heart disease (Taylor, 1972), the use of the two methods together is currently being examined (unpublished data).

CONCLUSIONS

In conclusion, it can be said that the CO_2 and N_2O rebreathing methods measure cardiac output reliably during exercise; the continuing use or introduction of these methods into routine hospital use is therefore recommended. Although there are limitations in each method, the roles of the two are complementary.

APPENDIX

Table A3.1: Values of mixed venous PCO_2, measured by various extrapolation procedures, in a group of subjects who rebreathed a range of CO_2 gas mixtures during exercise at a fixed work load. The oxygen consumption and cardiac output (based on Denison and Klausen $P\bar{v}CO_2$ estimates) are also shown for each subject.

T A B L E A 3.1: S T U D Y -1 and S T U D Y -2

SUBJECT	$\dot{V}O_2$ (ml/min)	Rebreathe mixture CO_2 %	Mixed venous PCO_2 (mmHg) estimated by the method of:						$\dot{Q}$ DENISON (l/min)	$\dot{Q}$ KLAUSEN (l/min)
			DENISON	KLAUSEN	KLAUSEN/ DEFARES	DEFARES	JERNERUS/ DEFARES	JERNERUS		
STUDY 1	1413	O	64	63	63	—	71	71	11.0	11.6
	1335	3	61	69	66	60.6	53	62	10.6	8.4
EZF	1280	6	62	74	74	—	—	—	10.3	7.6
	1273	8	59	58	58	58	60	60	11.3	11.6
	1195	P	60						10.3	
	1336	O	57	—	—	—	—	—	13.7	—
	1377	3	56	98	115	—	—	—	13.8	6.0
MR	1281	6	60	68	68	62	61	64	11.1	8.8
	1243	8	61	79	78	69	69	—	10.6	6.8
	1309	P	64						9.6	
	1279	O	64	86	86	—	—	—	10.2	6.2
	1292	3	64	99	99	—	—	—	10.6	5.1
PF	1260	6	66	92	92	95	—	84	10.0	5.7
	1234	8	66	66	67	65	67	67	10.1	9.9
	1292	P	69						9.4	
	1081	O	61	64	64	47	—	65	9.4	8.6
	1270	3	64	67	67	70	63	64	11.1	10.0
SS	1070	6	66	—	—	—	98	98	8.6	—
	1234	8	67	73	73	—	102	—	9.8	8.2
	1264	P	66						10.2	
	1311	O	66	74	74	—	60	59	12.6	9.4
	1284	3	67	66	66	65	64	67	13.4	14.0
JP	1261	6	63	85	85	78	73	66	13.2	7.1
	1250	8	61	—	—	—	—	—	13.2	—
	1246	P	65						12.2	

Table A3.1: continued.

SUBJECT	$\dot{V}O_2$ (ml/min)	Rebreathe mixture CO_2 %	Mixed venous PCO_2 (mmHg) estimated by the method of: DENISON	KLAUSEN	KLAUSEN/ DEFARES	DEFARES	JERNERUS/ DEFARES	JERNERUS	$\dot{Q}$ DENISON (l/min)	$\dot{Q}$ KLAUSEN (l/min)
JZ	1299	0	61	55	—	52	100	52	11.8	15.5
	1187	3	64	60	60	82	104	111	12.2	14.7
	1260	6	63	63	63	63	61	—	11.7	11.8
	1273	8	64	78	80	65	74	—	10.7	7.5
	1312	P	66						9.7	
HP	1226	0	63	56	56	54	53	51	11.0	14.2
	1222	3	62	70	70	62	64	64	11.6	8.9
	1284	6	63	66	66	63	60	58	11.6	10.5
	1237	8	65	65	65	65	85	86	11.0	11.0
	1184	P	68						9.4	
STUDY 2 EZF	1273	8	64	66	66	71	79	79	10.6	10.0
	1334	10	65	70	78	—	73	73	10.9	9.4
	1380	12	64	—	—	—	—	43	11.7	—
	1358	16	64	65	65	66	65	66	10.8	10.3
	1447	18	66	72	73	73	73	73	11.2	9.4
	1516	P	64						11.6	
MR	1342	8	63	62	62	62	63	63	14.7	15.7
	1314	10	62	69	—	59	66	66	13.6	10.6
	1342	12	64	63	63	61	63	63	13.2	13.2
	1348	16	66	70	69	70	70	70	12.6	9.5
	1266	18	68	71	72	72	72	72	12.4	11.2
	1317	P	65						12.3	
PF	1300	8	66	69	70	72	—	78	11.5	10.4
	1276	10	67	80	79	54	—	—	10.5	7.4
	1320	12	67	72	68	52	—	—	11.3	9.8
	1294	16	73	76	75	75	75	75	9.4	8.7
	1255	18	75	80	80	80	77	77	8.7	7.6
	1305	P	72						9.5	

Table A3.1: continued (2).

SUBJECT	$\dot{V}O_2$ (ml/min)	Rebreathe mixture CO_2 %	Mixed venous PCO_2 (mmHg) estimated by the method of: DENISON	KLAUSEN	KLAUSEN/ DEFARES	DEFARES	JERNERUS/ DEFARES	JERNERUS	$\dot{Q}$ DENISON (l/min)	$\dot{Q}$ KLAUSEN (l/min)
PK	1189	8	58	56	—	53	—	51	10.1	10.8
	1211	10	58	84	71	60	—	56	11.1	6.6
	1133	12	64	62	61	62	62	62	10.3	10.9
	1139	16	63	65	65	65	65	65	9.5	9.1
	1175	18	62	67	68	69	67	68	10.7	9.2
	1208	P	62						10.2	
ML	1148	8	58	—	50	50	50	78	13.0	—
	1181	10	59	65	61	61	63	—	12.8	11.6
	1148	12	61	58	59	59	59	—	11.2	12.4
	1211	16	63	66	67	67	68	75	12.2	10.8
	1204	18	63	71	71	71	71	77	11.4	8.9
	1192	P	64						11.5	
AA	1418	8	64	61	64	63	64	64	13.1	14.0
	1377	10	62	53	54	54	53	53	14.6	—
	1179	12	64	64	64	64	65	65	11.6	11.7
	1347	16	—	—	—	—	—	—	—	—
	1341	18	63	74	74	71	69	72	13.0	10.0
	1250	P	63						12.8	

Table A4.1: The mean values and coefficients of variation of repeated measurements of several parameters at rest and during exercise.

SUBJECT	WORK LOAD (watts)	No. of studies n	OXYGEN CONSUMPTION (ml/min) $\bar{m}$	CV%	CARBON DIOXIDE OUTPUT (ml/min) $\bar{m}$	CV%	MINUTE VENTILATION (l/min) $\bar{m}$	CV%	MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	CV%	CARDIAC OUTPUT (l/min) $\bar{m}$	CV%	HEART RATE (beats/min) $\bar{m}$	CV%	STROKE VOLUME (ml) $\bar{m}$	CV%
EZF 1	0		256	13.7	202	16.0	8.3	10.9	45.1	4.4	3.5	15.3	73	7.4	49	17.7
	50	15	965	5.8	819	6.5	24.3	5.4	60.7	2.9	8.3	7.4	97	7.3	86	11.0
	100		1572	7.6	1507	8.0	41.4	7.7	68.6	3.4	12.0	6.0	132	6.7	91	9.9
	150		2309	5.2	2338	6.3	64.6	4.2	74.3	2.9	15.6	3.1	173	4.8	90	5.5
MS 2	0		256	10.5	202	13.3	7.7	11.8	54.0	2.4	3.4	21.3	66	13.6	52	19.2
	50	10	1025	4.8	854	5.7	22.8	5.0	67.8	1.4	7.8	7.6	96	10.5	81	7.4
	100		1646	3.7	1503	5.1	37.5	5.4	74.5	0.9	11.3	4.0	127	8.6	90	6.6
	150		2363	4.2	2288	5.0	59.4	7.0	80.1	3.3	14.2	4.3	166	3.4	86	4.5
RD 3	0		243	11.5	202	13.3	7.9	15.9	45.0	4.8	5.1	6.6	87	8.5	59	22.5
	50	5	1106	4.3	960	5.8	28.9	5.7	60.4	1.6	10.0	2.9	110	5.0	91	4.3
	100		1640	3.1	1580	2.4	47.8	6.0	65.3	1.4	13.4	5.2	140	3.2	95	3.2
	150		2343	7.5	2412	6.8	74.4	5.9	68.1	0.5	17.3	2.5	167	3.5	104	4.8
PS 4	0		271	9.6	199	13.5	8.4	13.9	51.6	5.8	4.2	30.2	82	5.4	51	6.6
	50	6	1010	8.6	900	9.6	25.1	11.4	64.6	4.6	9.8	10.1	108	7.5	91	4.3
	100		1591	9.2	1646	2.7	42.7	8.9	68.4	3.3	14.6	4.2	146	7.7	101	4.5
	150		2120	10.0	2314	1.4	58.7	11.8	74.0	5.6	17.3	9.9	177	7.8	98	5.0
MT 5	0		257	10.1	228	6.7	7.2	5.8	48.4	5.1	3.9	19.9	62	11.4	62	24.4
	50	10	1034	9.2	888	8.3	24.5	8.0	65.8	4.4	8.1	9.0	83	7.7	99	4.8
	100		1584	5.3	1513	3.6	38.8	5.2	73.3	3.5	11.2	7.7	111	7.3	101	6.5
	150		2315	5.4	2338	5.0	57.8	5.3	79.3	3.0	15.0	5.0	143	3.2	105	4.6
ET 6	0		341	13.9	278	17.6	12.4	10.8	45.9	5.0	4.3	12.9	91	12.8	47	7.4
	40	6	928	5.8	870	10.1	28.3	6.3	56.0	2.5	9.8	5.2	123	6.3	80	4.7
	80		1383	3.4	1426	7.4	43.8	3.3	58.9	3.0	14.2	9.8	158	3.4	90	7.0
	120		1990	4.3	2120	6.4	67.8	4.5	61.9	2.7	17.9	4.3	184	2.1	97	3.3

Table A4.1: continued.

SUBJECT	WORK LOAD (watts)	No. of studies n	OXYGEN CONSUMPTION (ml/min) $\bar{m}$	CV%	CARBON DIOXIDE OUTPUT (ml/min) $\bar{m}$	CV%	MINUTE VENTILATION (l/min) $\bar{m}$	CV%	MIXED VENOUS PCO_2 (mmHg) $\bar{m}$	CV%	CARDIAC OUTPUT (l/min) $\bar{m}$	CV%	HEART RATE (beats/min) $\bar{m}$	CV%	STROKE VOLUME (ml) $\bar{m}$	CV%
AT 7	0	4	232	10.1	189	8.4	7.3	6.1	43.9	7.0	5.9	34.2	89	10.2	65	26.8
	35		846	7.2	762	6.6	25.1	2.4	57.4	2.0	8.4	6.6	125	1.8	67	5.7
	70		1205	5.0	1204	3.1	38.4	2.2	63.5	3.0	10.6	4.4	157	2.8	68	3.0
	105		1656	7.0			60.6	6.7	—	—	—	—	180	2.7	—	—
NT 8	0	4	264	12.3	170	14.9	8.1	3.6	47.5	3.5	4.0	23.9	98	12.6	41	29.3
	35		863	5.5	644	6.9	21.6	2.8	59.5	3.3	8.5	6.6	124	5.1	69	8.9
	70		1224	8.1	1136	3.6	30.7	4.7	65.7	1.5	9.5	3.5	151	4.9	63	4.9
	105		1723	4.7	1755	4.8	45.2	3.1	73.0	3.1	11.5	3.1	187	5.4	62	6.0
JZ 9	0	5	206	14.5	227	11.3	6.9	12.0	50.3	3.3	4.5	16.6	86	5.9	53	11.6
	35		727	4.3	756	5.8	19.4	7.3	61.1	3.1	7.5	9.0	119	3.7	59	12.3
	70		1199	4.0	1152	8.2	32.1	5.1	69.2	2.1	10.0	5.0	149	3.2	67	6.2
	105		1698	2.9	1747	4.6	50.5	5.4	77.9	1.2	12.1	8.0	184	1.5	66	9.3
SW 10	0	5	193	24.0	184	40.6	8.2	34.0	40.6	16.9	5.0	31.8	89	12.4	56	35.1
	33		763	2.4	580	9.6	18.7	5.6	57.8	4.6	6.6	5.1	128	3.3	52	7.6
	66		1200	2.6	1145	5.1	37.8	4.3	65.7	3.0	9.2	2.8	186	2.2	50	3.6

Table A5.1: Presenting the oxygen consumption, heart rate and cardiac output (indirect (CO_2) Fick method) in six subjects at three levels of work. Repeated measurements of cardiac output by the modified Cander and Forster, and Ayotte N_2O rebreathing methods are also shown, together with mean values and their coefficients of variation.

UBJ.	WORK (watts)	$\dot{V}O_2$ (ml/min)	HR (bts/min)	$\dot{Q}CO_2$ (l/min)	$\dot{Q}N_2O$ CANDER 1	2	3	4	$\bar{m}$	CV%	$\dot{Q}N_2O$ AYOTTE 1	2	3	4	$\bar{m}$	CV%
PF	40	828	86	7.45	7.81	—	5.06	8.64	7.17	—	—	—	—	—	—	—
	80	1256	103	9.65	5.30	9.50	8.08	8.76	7.91	23.1	5.67	9.85	5.83	6.50	6.91	28.0
	120	1876	136	13.40	15.22	15.71	12.42	12.83	14.05	11.8	9.22	9.21	12.79	—	10.41	—
GP	40	947	120	11.00	7.84	7.92	7.84	6.73	7.58	7.5	7.49	8.65	8.28	7.05	7.87	9.2
	80	1411	161	11.60	8.65	6.57	9.47	11.90	9.15	24.0	8.52	7.56	8.44	7.36	7.97	7.5
	120	1908	184	15.50	12.79	14.02	12.14	—	12.98	—	12.36	13.34	13.02	—	12.91	—
SS	40	947	94	10.00	9.95	9.34	9.25	7.52	9.01	11.5	7.78	7.55	6.82	4.60	6.69	21.7
	80	1424	111	10.50	8.61	8.45	7.20	10.32	8.65	14.8	9.30	8.24	9.26	9.71	9.13	6.9
	120	2094	141	14.00	11.46	14.05	12.50	13.06	12.77	8.5	10.32	8.80	10.24	10.65	10.00	8.2
MR	40	884	110	9.90	8.61	8.97	9.88	9.52	9.25	6.2	10.42	10.59	9.52	11.32	10.46	7.1
	80	1213	138	10.50	11.37	10.26	11.73	12.47	11.46	8.0	9.34	9.95	6.79	9.61	8.92	16.1
	120	1885	152	12.15	11.95	11.62	11.94	8.68	11.05	14.4	8.20	10.17	10.80	10.13	9.83	11.4
JP	40	1015	102	10.60	9.08	6.28	9.33	7.41	8.05	17.9	9.33	9.59	7.46	6.76	8.29	16.8
	80	1548	127	12.80	15.56	12.73	16.47	15.36	15.04	10.5	10.62	10.00	12.76	8.58	10.49	16.6
	120	1938	161	13.10	13.31	15.22	13.04	11.20	13.19	12.5	11.46	9.52	11.61	6.56	9.79	24.0
JJ	40	945	137	10.80	10.26	9.46	8.32	8.23	9.07	10.7	8.27	6.91	8.10	7.52	7.70	8.1
	80	1421	147	11.40	12.71	10.13	10.34	9.14	10.58	14.3	6.89	7.41	7.85	9.16	7.83	12.4
	120															
PK	40	778	116	7.90	5.55	8.49	8.44	6.74	7.31	19.6	6.21	7.50	4.16	6.47	6.09	23.0
	80	1246	135	9.70	9.45	9.36	9.15	8.58	9.14	4.3	6.67	5.82	6.59	5.96	6.26	6.9
	120	2021	177	12.60	11.23	11.08	10.98	11.39	11.17	1.6	10.67	9.23	11.02	11.57	10.62	9.4

Table A6.1: Repeated measurements of oxygen consumption, cardiac output, heart rate and stroke volume are shown in nine subjects at work loads of 300, 600, and 900 kpm/min. The mean value of each parameter, and its coefficient of variation is presented for each subject, and at each work load.

SUBJECT		WORK 300 kpm/min							600 kpm/min							900 kpm/min						
							$\bar{m}$	CV%						$\bar{m}$	CV%						$\bar{m}$	CV%
MM	$\dot{V}O_2$	1092	1071	1060	1004	1096	1065	3.5	1646	1643	1642	1647	1738	1663	2.5							
	$\dot{Q}$	11.4	12.0	12.0	11.7	11.7	11.8	2.1	15.6	16.3	16.7	17.7	16.9	16.7	4.1							
	HR	90	86	92	91	89	90	2.5	103	105	114	116	116	111	5.7							
	SV	127	139	130	129	132	131	3.5	152	155	147	153	146	151	2.6							
RHTE	$\dot{V}O_2$	773	790	782	781	745	782	1.0	1221	1182	1180	1223	1192	1198	1.6							
	$\dot{Q}$	9.1	7.2	7.7	8.1		8.0	10.3	13.0	12.4	11.3	13.7	11.2	12.3	3.9							
	HR	78	77	78	79		78	1.1	95	100	98	100	100	99	2.2							
	SV	117	93	98	103		103	10.0	136	124	115	137	112	125	9.3							
GH	$\dot{V}O_2$	909	920	953	936	877	919	3.1	1412	1418	1382	1401	1428	1408	1.3	2057	2326	2261	2237	2271	2230	4.6
	$\dot{Q}$	9.3	11.3	8.6	10.0	10.7	9.9	10.9	12.2	13.2	14.9	13.2	11.7	13.1	9.6	18.8	19.9	20.3	20.6	19.9	19.9	3.5
	HR	92	90	91	95	95	93	2.4	120	127	129	128	129	127	3.0	150	158	162	164	163	159	3.6
	SV	101	125	94	105	112	107	11.0	101	104	116	103	91	103	8.6	125	126	125	126	122	125	1.3
DN	$\dot{V}O_2$								1326	1396	1357	1489	1494	1412	5.4	1946	1957	1987	2012	2021	1985	1.7
	$\dot{Q}$								12.1	11.4	12.1	14.4	11.4	12.3	9.9	17.2	17.6	19.7	17.8	18.7	18.2	5.6
	HR								116	119	118	123	121	119	2.3	122	128	135	134	140	132	5.2
	SV								104	95	102	117	94	102	8.9	141	137	146	133	133	138	4.1

Table A6.1: continued.

SUBJ.		WORK 300 kpm/min					$\bar{m}$	CV%	600 kpm/min					$\bar{m}$	CV%	900 kpm/min					$\bar{m}$	CV%
EZF	$\dot{V}O_2$	889	857	868	847	864	865	1.8	1414	1454	1636	1496	1386	1477	6.6	1989	1844	2076	1961	2213	2017	6.2
	$\dot{Q}$	11.5	11.8	10.1	12.1	10.6	11.2	4.7	14.2	14.3	15.1	15.9	15.3	14.9	4.8	15.7	14.8	16.5	16.4	17.2	16.1	5.5
	HR															130	153	157	157	159	151	7.9
	SV															121	98	105	104	108	107	7.9
JR	$\dot{V}O_2$	865	866	865	857	903	871	2.1	1253	1261	1503	1298	1420	1347	8.2	2390	2340	2403	2478	2514	2425	2.9
	$\dot{Q}$	8.2	9.0	8.9	8.8	8.9	8.7	3.7	11.5	13.3	14.1	12.8	13.4	13.0	7.5	15.3	14.7	14.7	16.6	16.5	15.5	6.1
	HR	90	98	91	94	97	94	3.7	116	115	117	118	118	117	1.1	148	151	158	162	167	157	5.0
	SV	91	92	98	93	92	93	3.0	99	116	120	109	113	111	7.2	103	97	93	102	99	99	4.0
PH	$\dot{V}O_2$	878	732	786	828	703	785	9.0	1334	1382	1418	1383	1388	1381	2.2	2141	2093	2228	2061	2617	2228	10.1
	$\dot{Q}$	9.5	11.0	10.7	11.1	11.6	10.8	7.2	14.3	15.3	15.8	15.7	15.4	15.3	3.8	16.7	16.0	19.0	17.8	17.2	17.3	6.5
	HR	109	108	109	111	111	110	1.2	131	134	138	137	143	137	3.3	175	182	188	187	189	184	3.2
	SV	87	102	98	100	104	98	6.6	109	114	114	115	107	112	3.2	95	88	101	95	91	94	5.2
PS	$\dot{V}O_2$	792	880	784	839	924	844	7.0	1538	1570	1643	1614	1571	1587	2.6	1983	2349	2244	2707	2307	2221	7.4
	$\dot{Q}$	7.9	9.6	10.8	7.6	10.6	9.3	16.1	12.0	13.1	11.9	10.5	13.8	12.3	10.4	15.2	17.4	15.5	15.6	—	16.0	6.1
	HR	115	116	122	121	125	120	3.5	151	161	166	165	163	162	2.7	179	188	168	183	187	181	4.5
	SV	69	82	88	63	85	77	14.3	77	82	72	64	85	76	10.9	85	93	92	85	109	93	10.6

Table A6.1: continued (2).

SUBJ.		WORK 300 kpm/min					$\bar{m}$	CV%	600 kpm/min					$\bar{m}$	CV%	900 kpm/min				$\bar{m}$	CV%
DB	$\dot{V}O_2$								1401	1399	1494	1526	—	1455	4.5	1993	2299	2103	2063	2115	6.2
	$\dot{Q}$								13.7	12.9	12.0	12.8		12.8	5.1	19.5	17.9	19.5	17.3	18.5	6.1
	HR								95	96	99	102		98	3.2	136	138	146	150	143	4.6
	SV								144	134	122	125		131	7.6	143	129	134	115	130	9.0

Table A7.1: Repeated measurements of oxygen consumption and cardiac output (using the indirect (CO_2) Fick method) in nine subjects at work loads of 300, 600 and 900 kpm/min. Two values of cardiac output are given; $\dot{Q}CO_2$ "corr" values are based on an estimate of $P\bar{v}CO_2$ corrected for the "downstream" effect, while $\dot{Q}CO_2$ "uncorr" values are based on the measured $PvCO_2$. The mean value of each parameter, and its coefficient of variation is presented for each subject, and at each work load.

SUBJ.		WORK LOAD: 300 kpm/min							600 kpm/min							900 kpm/min						
							$\bar{m}$	CV%						$\bar{m}$	CV%						$\bar{m}$	CV%
MM	$\dot{V}O_2$	1015	1074	1045	1043	1108	1057	3.3	1647	1608	1735	1692	1732	1683	3.3							
	Corr. $\dot{Q}CO_2$	8.9	11.6	10.7	10.2	11.5	10.6	10.3	12.8	14.6	15.5	14.6	15.1	14.5	7.1							
	Uncorr. $\dot{Q}CO_2$	7.8	9.8	9.5	9.2	10.3	9.3	10.0	10.5	11.9	12.8	12.3	12.8	12.1	7.9							
RHTE	$\dot{V}O_2$	749	692	772	732	793	747	5.1	1247	1219	1282	1257	1322	1265	3.1							
	Corr. $\dot{Q}CO_2$	11.8	10.6	10.5	9.1	9.4	10.3	10.5	13.8	12.9	14.1	13.5	13.8	13.6	3.3							
	Uncorr. $\dot{Q}CO_2$	9.5	8.5	8.7	7.4	7.8	8.4	9.7	10.8	10.2	11.1	10.6	11.3	10.8	4.0							
GH	$\dot{V}O_2$	840	958	811	871	975	891	8.1	1356	1464	1431	1399	1382	1406	3.0	1840	2231	2329	2229	2473	2220	1.1
	Corr. $\dot{Q}CO_2$	9.4	10.0	8.6	9.4	9.6	9.4	5.4	17.7	18.2	15.5	15.6	16.0	16.6	7.6	19.0	18.1	19.1	20.5	19.3	19.2	4.5
	Uncorr. $\dot{Q}CO_2$	7.7	8.4	7.3	8.0	8.1	7.9	5.3	14.1	14.7	12.6	12.8	13.1	13.5	6.7	15.3	15.0	15.9	17.2	16.3	15.9	5.5

Table A7.1: continued.

UBJ.		WORK LOAD: 300 kpm/min					$\bar{m}$	CV%	600 kpm/min					$\bar{m}$	CV%	900 kpm/min					$\bar{m}$	CV%
DN	$\dot{V}O_2$								1402	1274	1399	1526	1458	1412	6.6	1913	1968	2024	1946	1904	1951	2.5
	Corr. $\dot{Q}CO_2$								15.2	16.2	19.6	20.1	19.6	18.1	12.4	18.8	20.2	17.8	20.1	20.1	19.4	5.5
	Uncorr. $\dot{Q}CO_2$								12.0	13.0	15.5	15.9	15.5	14.4	12.2	14.7	15.9	14.2	16.0	16.1	15.4	5.7
ZF	$\dot{V}O_2$	892	878	873	899	901	888	1.5	1352	1413	1331	1483	1407	1397	4.2	1782	2004	2043	2027	2147	2000	6.7
	Corr. $\dot{Q}CO_2$	13.2	10.8	11.3	12.0	12.4	11.9	7.9	17.9	18.9	17.4	17.2	16.3	17.6	5.2	20.2	20.9	19.3	22.8	20.0	20.6	6.4
	Uncorr. $\dot{Q}CO_2$	10.2	8.9	9.0	9.7	10.1	9.6	6.3	13.8	14.6	13.8	13.9	13.1	13.8	3.8	16.2	16.8	15.6	18.4	16.4	16.7	6.3
JR	$\dot{V}O_2$	829	796	857	837	—	830	3.0	1229	1152	1459	1305	—	1286	10.2	2263	2472	2391	2604	2712	2488	7.1
	Corr. $\dot{Q}CO_2$	10.0	10.2	9.8	8.4	—	9.6	8.4	11.4	11.2	14.7	12.9	—	12.6	12.7	21.7	17.9	20.0	19.8	18.3	19.5	7.7
	Uncorr. $\dot{Q}CO_2$	8.1	8.3	7.9	6.9	—	7.8	7.9	9.1	8.9	11.7	10.6	—	10.1	12.8	16.4	14.4	15.5	16.1	15.0	15.5	5.2
PH	$\dot{V}O_2$	855	925	809	824	—	853	6.0	1307	—	1372	1396	1429	1376	3.8	2015	2131	2212	1975	2304	2127	6.4
	Corr. $\dot{Q}CO_2$	11.7	15.0	12.2	11.7	—	12.7	12.5	13.9	—	14.2	15.3	17.4	15.2	10.5	18.8	18.2	16.9	15.2	17.0	17.2	8.0
	Uncorr. $\dot{Q}CO_2$	9.7	11.5	9.8	9.4	—	10.1	9.4	11.0	—	11.3	12.2	13.7	12.1	10.0	15.4	15.1	14.1	12.5	14.2	14.7	13.0

Table A7.1: continued (2).

SUBJ.		WORK LOAD: 300 kpm/min					$\bar{m}$	CV%	600 kpm/min					$\bar{m}$	CV%	900 kpm/min					$\bar{m}$	CV%
PS	$\dot{V}O_2$	930	748	918	830	—	857	9.9	1630	—	1646	1534	1637	1612	3.2	2157	2045	2513	2530	—	2311	10.7
	Corr. $\dot{Q}CO_2$	9.8	10.9	11.0	13.5	—	11.3	8.1	16.2	—	15.4	14.1	15.8	15.4	14.1	21.0	20.5	19.0	21.7	—	20.6	5.5
	Uncorr. $\dot{Q}CO_2$	8.2	8.7	9.0	11.0	—	9.2	13.1	13.2	—	12.3	11.4	12.8	12.4	6.3	16.0	15.9	15.9	17.7	—	16.4	5.4
DB	$\dot{V}O_2$															2189	1997	2064	2168	—	2105	4.3
	Corr. $\dot{Q}CO_2$															19.2	17.2	19.3	20.7	—	19.1	7.5
	Uncorr. $\dot{Q}CO_2$															15.3	13.8	15.2	16.6	—	15.2	7.5

Table A8.1: A list of the studies which provided the "collected data" for dye dilution and direct (O_2) Fick estimates of cardiac output quoted in the thesis.

METHOD	TYPE of EXERCISE	No. of OBSERVATIONS	SOURCE
DYE DILUTION	BICYCLE SITTING POSITION	209	Astrand, et al. 1964 Grimby, et al. 1966 Hermansen, et al. 1970 Stenberg, et al. 1967
DIRECT (O_2) FICK	BICYCLE SITTING POSITION	56	Bevegard, et al. 1960 Bevegard, et al. 1963 Bevegard, et al. 1966 Ekelund & Holmgren 1967 Ekelund 1967

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