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Impact of Exercise Duration on Maximal and Sub-Maximal Markers during Clinical Cardio-Pulmonary Exercise Testing

By

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A thesis submitted in fulfilment of the degree of Doctor of Philosophy (PhD) of exercise physiology to the College of Medical, Veterinary and Life Sciences, in the University of Glasgow

Submission date February 2012



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GLASGOW**

Author's declaration

I hereby declare that this thesis was composed by myself and that all the work reported was performed by myself except where assistance has been acknowledged, where other sources of information have been used.

Signature:

Date:

Abstract

Currently, the American College of Sports Medicine (ACSM) recommends that protocols for cardiopulmonary exercise testing (CPET) should last between eight and twelve minutes. However, the justification for these exercise durations rely on limited experimental data. These recommendations have a significant impact on the ability of frail patients to be assessed using CPET and should conform to evidence based practice. This thesis begins by assessing the validity of these recommendations in relation to maximal exercise responses before assessing the consequences of these recommendations on sub-maximal exercise measurements. These studies were conducted in a relatively large cohort (compared to the study that underpins the ACSM guidelines) of heterogeneous volunteers (they are both men and women, with a significant age range and varied functional capacity) to make the data more relevant to clinical exercise testing.

The data presented in chapter three demonstrate that it is very difficult to obtain exercise duration conforming to the current ACSM guidelines by using a standardised ramp exercise protocol on both treadmill and cycle ergometer exercise. However, sub-group analyses for those subjects who achieved moderate (8-12 minutes) and short (less than 8 minutes) exercise durations. In addition, a separate analysis was carried out for a different sub-group of those who achieved moderate (8-12 minutes) and long (more than 12 minutes) of durations of exercise. Despite this, it was possible to demonstrate in sub-group analysis that there was no significant difference in peak oxygen uptake, peak carbon dioxide output, peak heart rate, peak ventilation and peak power output when exercise duration was less or more than that prescribed by the ACSM recommendations. In addition, the effects of long, moderate or short duration exercise *per se* were also analysed in this chapter and again exercise duration was shown to be without effect on the main maximal markers of exercise performance.

In chapters four, five and six, the initial findings were extended to determine the effects of exercise duration on a range of clinically relevant sub-maximal markers of exercise performance. It was likely, since exercise duration did not

affect maximal exercise that the physiological determinants of maximal performance were not significantly altered during short or long duration exercise and consequently it was likely that sub-maximal markers of functional capacity would not be affected. However, the quality of the data obtained during CPET can obviously influence the accurate measurement physiological responses during exercise and much of the analysis in these chapters focused on the validity of the data analysis.

Chapter four investigated the limitations to measuring the break point in the relationship between oxygen uptake and carbon dioxide output during progressive exercise (the so called ventilatory threshold or 'V-slope'). The accurate measurement of this break point was determined by standard gas exchange criteria and the effects of reducing the data available for analysis (by reducing the amount of breaths available for comparison at reduced exercise durations) were examined. The data showed that reducing the data available for analysis had an impact on the quality of the data (decreasing the goodness of fit) but no significant effect on the determination of the ventilatory threshold.

Chapter five determined the effects of exercise duration on the oxygen uptake efficiency slope (OUES). As expected, the effects of exercise duration were not significant but additional investigation into the commonly employed data analysis procedures was performed. These data show that the log transformation of the relationship between ventilation and oxygen uptake allows reliable assessment of ventilatory efficiency in most cases, however, the impact of the lactate threshold on ventilation and the biological variability in where the threshold occurs as a proportion of functional capacity can impact on the sensitivity of this measurement to predict aerobic and/or anaerobic capacity.

Chapter six determined the effects of exercise duration on the breathing reserve index and found no significant difference during short, moderate or long exercise duration exercise. Further analysis was performed to demonstrate limitations in the use of predicted maximum voluntary ventilation (rather than direct measurement).

Taken together, these data demonstrate that the current ACSM recommendations for CPET are too restrictive and may limit the application of

such testing in populations that cannot exercise for between eight and twelve minutes. The data further suggest that the testing and analysis procedures used during CPET are central to producing valid maximal and sub-maximal markers of functional capacity and the recommendations should focus include guidelines in relation to such aspects.

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I would like to thank my supervisor, Dr Niall G MacFarlane, for the patient guidance, encouragement and advices. I would like to express my deep appreciation to my volunteers. I would also like to thank all the members of staff at the University of Glasgow. I would like to thank Mr John Wilson for his very helpful comments, suggestions, improvements, and corrections to the cardiopulmonary exercise testing. There are a few people, I would like to thank individually; I would like to thank Dr Nazim Ghouri for clinical examination of subjects before scheduling them to maximal exercise testing. I would like to thank Mr Paul Paterson for his support and help.

The last thank you goes to my family; I would like to thank my wife and sons for being the ultimate reason for finishing this thesis and for their love and support.

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Figure 5-6 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{\text{before}}$ including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-7 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{\text{before}}$ excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-8 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{\text{after}}$ including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-9 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{\text{after}}$ excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-10 Pearson's correlation coefficient measures the linear relations between $OUES$ and \dot{V}_{O_2} (ml/min), during moderate treadmill studies. For both male and female as one group and including cardio-dynamic phase.

Figure 5-11 illustrates total oxygen uptake efficiency slope $OUES_{\text{Total}}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-12 illustrates total oxygen uptake efficiency slope $OUES_{\text{Total}}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-13 illustrates 75% of oxygen uptake efficiency slope $OUES_{75\%}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-14 illustrates 75% of oxygen uptake efficiency slope $OUES_{75\%}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-15 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{\text{before}}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-16 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{\text{before}}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-17 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{\text{after}}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-18 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{\text{after}}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5-19 Pearson's correlation coefficient measures the linear relations between $OUES$ and \dot{V}_{O_2} (ml/min), during moderate cycle ergometer studies. For both male and female as one group and including cardio-dynamic phase.

Figure 6-1 illustrates peak breathing reserve index (BRI_{Peak}) during short moderate and long treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6-2 illustrates peak breathing reserve index at anaerobic threshold point (BRI_{AT}) during short moderate and long treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6-3 Correlation between (BRI_{Peak}) and (BRI_{AT}) during short, moderate and long exercise duration studies and for all data together during treadmill studies.

Figure 6-4 illustrates peak breathing reserve index (BRI_{Peak}) during short moderate and long cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6-5 illustrates peak breathing reserve index at anaerobic threshold point (BRI_{AT}) during short moderate and long cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6-6 Correlation between (BRI_{Peak}) and (BRI_{AT}) during short, moderate and long exercise duration studies and for all data together during cycle ergometer studies. NS= no significant

Figure 6-7 Correlation of (A) (BRI_{Peak}) and (B) (BRI_{AT}) with $\dot{V}_{O_2 \text{ PEAK}}$ for all data together during treadmill studies, NS = no significant correlation.

Figure 6-8 Correlation of (A) (BRI_{peak}) and (B) (BRI_{AT}) with $\dot{V}_{O_2 PEAK}$ for all data together during cycle ergometer studies, NS = no significant correlation.

Figure 6-9 demonstrates that (A), (BRI_{AT}) is not different during treadmill and cycle ergometer studies (0.25 ± 0.05 in treadmill 0.23 ± 0.05 in cycle ergometer for all volunteers, $P=NS$), whereas (BRI_{peak}) is marginally higher during in treadmill studies but not significantly (0.69 ± 0.11 in treadmill vs 0.63 ± 0.15 in cycle ergometer for all volunteers, $P=NS$).

Figure 6-10 demonstrates that there is no real agreement between (BRI_{AT}) and ventilatory anaerobic threshold during the test ramp protocols on treadmill (A) and cycle ergo meter (B).

Definitions of abbreviations

ACSM	American College of Sports Medicine	
B_f	Breathing frequency	(Breaths/min)
BRI_{peak}	Peak breathing reserve index	
BRI_{AT}	Breathing reserve index at	
AT	Anaerobic threshold	
BSA	Body Surface Area	m^2
CAD	Coronary artery disease	
CHD	Coronary heart disease	
CHF	Congestive heart failure patients	
COPD	Chronic obstructive pulmonary disease	
CO_2	Carbon dioxide product	
CPET	Cardiopulmonary exercise testing	
$F_{ET}CO_2\%$	fraction of end tidal CO_2 concentration	
$F_{ET}O_2\%$	fraction of end tidal O_2 concentration	
HR	Heart rate	$beat \cdot min^{-1}$
HR_{max}	Maximal heart rate	$beat \cdot min^{-1}$
HR_{peak}	peak heart rate	$beat \cdot min^{-1}$
LT	Lactate threshold	

MVV	Maximum voluntary ventilation	(l/min)
OUES	Oxygen uptake efficiency slope	
PVD	peripheral vascular disease	
Q	Cardiac output	
RER	Respiratory gas exchange ratio	
RCP	Respiratory compensation point	
RQ	Respiratory quotient	
RSS	residual sum of square	
%SaO ₂	saturation of arterial blood	
VAT	$\dot{V}O_2$ at anaerobic threshold	l.min ⁻¹
\dot{V}_{CO_2}	carbon dioxide production	l.min ⁻¹
\dot{V}_E	Minute ventilation	l.min ⁻¹
$\dot{V}_{O_2 \text{ PEAK}}$	Peak oxygen consumption	l.min ⁻¹
$\dot{V}_{O_2 \text{ MAX}}$	Maximal oxygen consumption	l.min ⁻¹
V _t	Tidal volume	l
WR	Workload	watt/min
WR _{Peak}	Peak Workload	watt

Definitions of Units

α	the angle of inclination in degrees
Adjusted R^2	adjusted square of correlation coefficient
BMI	body mass index
$b.m^{-1}$	beat per minute
CI	confidence interval
g	standard gravity $9.81m.s^{-2}$
kg	kilogram
l/min	litter per minutes
m	body mass in kilogram
$ml^{-1}.kg^{-1}.min^{-1}$	millilitre per kilogram every one minutes
P	level of significant
r	Pearson's correlation coefficient
R^2	square of correlation coefficient
rpm	revolution per minutes
S	seconds
SD	standard deviation
V(t)	time of velocity in seconds
W/min	Watt per minutes

CHAPTER ONE

Introduction

1 Introduction

1.1 Cardio-Pulmonary Exercise Testing

The physiological data generated during cardiopulmonary exercise testing (CPET) can be used to determine the aetiology of the factors limiting exercise performance. Consequently, the performance of (CPET) valid and reproducible procedures that ensure the data generated can accurately reflect ongoing physiological processes. Maximal and sub-maximal data can be important for understanding exercise performance and this thesis will address issues relating to the generation of both. Detailed discussion of the physiological basis for the chosen performance markers will be reserved to the introduction for the relevant data chapters and the following general introduction will only provide an overview of the relevant literature and rationale for the programme of scientific study that was developed.

Cardiopulmonary exercise testing is now being utilised more than ever before (Pina *et al.*, 1995;ATS/ACCP, 2003;Lear *et al.*, 1999;Franklin *et al.*, 2000). It is commonly used as a non-invasive procedure to assess functional capacity in patients suffering from heart and lung disorders (Grant *et al.*, 1998;Lipkin *et al.*, 1986;Revill *et al.*, 2002;Patterson *et al.*, 1972) and to assess the safety of heart transplantation (Mancini *et al.*, 1991). In addition, (CPET) can be employed to study the effect of medical treatments in a range of clinical populations (Mezzani *et al.*, 2009;Bjornstad H, 2001) and can also be used to determine functional capacity and the effects of exercise training in healthy populations (Mollard *et al.*, 2008;Akkerman *et al.*, 2010).

1.2 Safety and risk during Cardio-Pulmonary Exercise Testing

Even in clinical situations (CPET) is relatively safe and this is particularly so when patients have a low risk of coronary heart disease (CHD) (McHenry, 1977;Gibbons *et al.*, 1989). According to previous reports, serious complications requiring hospitalisation are less than 5 in 10,000 tests and mortality during (CPET) is 0.5 in 10,000 tests (Stuart Jr & Ellestad, 1980;Gibbons *et al.*, 1989).

Even in heart failure patients the risk is relatively low, with no deaths recorded in a series of 4,411 (CPETs) from 2,037 heart failure patients and an incidence of non-serious complications of only 0.5 in 1,000 (Keteyian *et al.*, 2009).

It is seldom that sudden cardiac death occurs in healthy individuals less than 40 years of age (Fletcher *et al.*, 2001) and similarly the incidence of sudden death during (CPET) in patients with coronary arteries disease (CAD) was estimated at 1 in 80,000-160,000 (CPET) hours (Cobb & Weaver, 1986).

1.3 Measurements during Cardio-Pulmonary Exercise Testing

Cardio-pulmonary exercise testing with open-circuit spirometry to the limit of exhaustion or symptoms is widespread in clinical practice. In practice, the patient inhales ambient air (normally with an O₂ concentration of 20.93% and a CO₂ concentration of 0%) and the expired gas volume is measured and a sample tested by sensitive O₂ and CO₂ analysers to determine changes in the concentration of these gases.

Normally, there is a significant physiological reserve in cardio-pulmonary system at rest and any physiological work that increases the metabolic rate will require a rapid cardio-pulmonary response. The importance of (CPET) is, therefore, to assess the physiological response to increasing metabolic load and ensure that the cardio-pulmonary system responds quickly and appropriately to support full functional capacity in healthy (Mitchell & Blomqvist, 1971) or sporting populations (Saltin & Astrand, 1967) and to help assess the nature of the cardio-pulmonary impairment limiting exercise capacity in patient populations (Mancini *et al.*, 1991).

Therefore, the testing procedure commonly involves the measurement of oxygen uptake at the mouth \dot{V}_{O_2} , carbon dioxide output at the mouth \dot{V}_{CO_2} , minute ventilation \dot{V}_E and heart rate HR during graded incremental exercise testing or ramp exercise testing protocols (ATS/ACCP, 2003; Lear *et al.*, 1999; Franklin *et al.*, 2000).

Hill and colleagues were the first scientists to introduce maximal oxygen uptake $\dot{V}_{O_2 \text{ MAX}}$ as a measurement for endurance capacity (Hill & Lupton, 1923) and even

today $\dot{V}_{O_2 \text{ MAX}}$ is one of the most common physiological measurements made in exercise physiology laboratories. In addition to the term maximum oxygen consumption the measurement is also often referred to as maximum oxygen uptake, maximum oxygen power, exercise capacity, exercise tolerance and functional capacity (Meyer *et al.*, 2005; Balady *et al.*, 2010). No matter what it is called, the measurement represents the ability of human body to take oxygen from the respiratory system, transport that oxygen through the cardiovascular system and finally use it to support energy production in active muscles (Shephard, 1984; Yoon *et al.*, 2007; Carlson, 1995) .

$\dot{V}_{O_2 \text{ MAX}}$ is a good indicator for cardiopulmonary fitness (Carlson, 1995) and a marker of functional capacity in the assessment of many clinical conditions (Franklin *et al.*, 2000; Lear *et al.*, 1999). It has also been used to express patient's ability to perform sub-maximal activities *e.g.* in a 6-minute walk test (Alameri *et al.*, 2009) and the prognostic assessment of heart disorders. In large sample studies, a low $\dot{V}_{O_2 \text{ MAX}}$ (less than 10 ml/kg/min) indicates a poor prognosis whereas a higher value (over 18 ml/kg/min) suggests better prognosis (Opasich, 1998).

(Mancini *et al.*, 1991) reported that when $\dot{V}_{O_2 \text{ MAX}}$ is used to assess functional capacity during pre-operative investigation for cardiac transplantation then a threshold of 14 ml/kg/min should be used to determine when the transplant was imminently necessary.

1.4 The physiological response to exercise

The transfer of oxygen from the atmosphere in the respiratory system and its transport via the cardiovascular system and consumption within active muscle are simultaneous and interrelated processes. Cardiopulmonary exercise testing provides a mechanism to investigate the physiological response to increasing workload and to establish the relative efficacy of the main physiological systems in supporting the demands of the exercise.

$\dot{V}_{O_2 \text{ MAX}}$ can be defined by the Fick equation as

$$\dot{V}_{O_2 \text{ MAX}} = Q (CaO_2 - CvO_2) \quad (\text{equ.1.1})$$

where (Q) is the cardiac output (the product of heart rate and stroke volume), CaO_2 is the arterial oxygen content and CvO_2 the venous oxygen content obtained at maximal effort.

This definition clearly illustrates the interrelationship between central haemodynamics and peripheral muscle function and as such the main physiological factors that influence $\dot{V}_{O_2 \text{ MAX}}$ can be considered be either 'central' or 'peripheral'.

1.5 Central factors limiting $\dot{V}_{O_2 \text{ MAX}}$

1.5.1 Cardiac output

Hill identified maximal cardiac output (Q) as the principle factor for individual differences in $\dot{V}_{O_2 \text{ MAX}}$ and since maximal heart rate does not show a considerable variation in people of the same age any difference in $\dot{V}_{O_2 \text{ MAX}}$ has been closely related to maximum stroke volume (Bassett, Jr. & Howley, 2000).

McArdle and colleagues studied exercise response in three groups and compared heart rate, stroke volume, cardiac output and $\dot{V}_{O_2 \text{ MAX}}$ (McArdle *et al.*, 1991). The three groups were athletes, sedentary subjects and patients with mitral valve stenosis. Maximum heart rate was similar in all groups but cardiac output was highest in athletes as a result of their large stroke volume. The mitral stenosis patients had the lowest stroke volume and consequently the lowest $\dot{V}_{O_2 \text{ MAX}}$. The group of athletes had, on average, a 62.5% higher $\dot{V}_{O_2 \text{ MAX}}$ than those that were sedentary and this observation paralleled, on average, a 60% higher stroke volume.

Similar results were found in a further study where sedentary volunteers were subjected to an 8-week aerobic training intervention that demonstrated a 35% increase in stroke volume and a similar magnitude increase in cardiac output

(McArdle *et al.*, 1991). With further work by (Saltin & Strange, 1992) showing that aerobic training after enforced bed rest resulted in an increase in $\dot{V}_{O_2 \text{ MAX}}$ due predominantly to increases in cardiac output. In fact, since little O_2 can be extracted from the arteries blood at maximal exercise it suggests that the main determinant of $\dot{V}_{O_2 \text{ MAX}}$ is related to O_2 delivery *i.e.* to cardiac output. It has been estimated that 70-85% of the limitation to $\dot{V}_{O_2 \text{ MAX}}$ is centrally linked to maximum cardiac output (Bassett, Jr. & Howley, 2000).

Thus (CPET) is very sensitive to changes in central cardiac haemodynamics and can provide a sensitive and reliable non-invasive assessment of cardiovascular function to help in the determining the aetiology of symptoms, the impact of novel therapeutic regimens and prognosis.

1.5.2 Pulmonary diffusion capacity

The saturation of arterial blood (%SaO₂) remains fairly constant above 95% at rest and during maximal effort (Powers *et al.*, 1989) and suggests a less critical role for pulmonary diffusion to limit $\dot{V}_{O_2 \text{ MAX}}$. However, the impact of reduced pulmonary diffusion on $\dot{V}_{O_2 \text{ MAX}}$ can be easily observed as a reduction in exercise capacity at high altitude (*e.g.* between 3000-5000 m) (Faulkner *et al.*, 1968; Daniels & Oldridge, 1970). The reduced exercise capacity reflects reduced oxygen levels in this hypobaric environment and consequently a reduced driving force for gas exchange across the alveolar membrane. Conversely, exercise performance can be enhanced by increasing the driving force for pulmonary diffusion though breathing hyperoxic gas mixtures under experimental conditions (Powers *et al.*, 1989).

These observations suggest that pulmonary diffusion can play a minor role in limiting $\dot{V}_{O_2 \text{ MAX}}$ under extreme environmental conditions. However, desaturation of arterial blood can be observed in extremely well trained athletes at high cardiac outputs (Dempsey *et al.*, 1984). These high cardiac outputs reduce blood transit time across the pulmonary circulation and limit the capacity for O_2 to diffuse across the alveolar membrane. Similar desaturation can also be observed in patient populations (*e.g.* in COPD and exercise induced asthma) (Rooyackers

et al., 1997). In such situations, the use of (CPET) can be instrumental in identifying pulmonary limitations as a mechanism for the exercise intolerance.

1.5.3 O₂ carrying capacity

Haemoglobin (Hb) is an iron containing metalloprotein that in red blood cells. It has an oxygen binding capacity of 1.34ml O₂/g Hb that increases the amount of O₂ carrying capacity of the blood by a factor of 70 (over oxygen dissolved in plasma). Increasing the O₂ carrying capacity, e.g. by blood doping where red blood cells are removed, stored and later infused to artificially increase the total Hb levels by as much as 8-20%, can improve $\dot{V}_{O_2 \text{ MAX}}$ by 4-9% (Bassett, Jr. & Howley, 2000; McArdle *et al.*, 1991). In conditions, such as anaemia, there is a linear fall in $\dot{V}_{O_2 \text{ MAX}}$ with Hb levels (Lindstedt & Wells, 1988) and again (CPET) is useful in identifying the cause of exercise intolerance of symptoms of breathlessness or fatigue.

1.6 Peripheral factors limiting $\dot{V}_{O_2 \text{ MAX}}$

1.6.1 Peripheral diffusion gradients

In the skeletal muscle, the main resistance to O₂ diffusion is at the interface between the capillary wall and the sarcolemmal membrane (Honig *et al.*, 1992). In animal studies, Grassi established that difference between capillary and intracellular Po₂ was the main factor in supporting O₂ conductance across the cell membrane (Grassi, 2000). Under normal circumstances, the increase in skeletal muscle blood flow with exercise and increased tissue metabolism will support the necessary Po₂ gradient and mean that peripheral diffusion does little to limit $\dot{V}_{O_2 \text{ MAX}}$. However, peripheral vascular disease will reduce tissue perfusion and the capillary sarcolemmal pO₂ gradient and may limit $\dot{V}_{O_2 \text{ MAX}}$.

1.6.2 Mitochondrial factors

Under normal circumstances, oxygen consumption by the mitochondrial electron transport chain will determine oxygen uptake at the lungs. Even so, increasing mitochondrial activity by a factor of 2 will only increase $\dot{V}_{O_2 \text{ MAX}}$ by 20-40% (Bassett,

Jr. & Howley, 2000; Holloszy & Coyle, 1984). Increasing mitochondrial activity may relate to increase in mitochondrial density (increasing volume or number) or up-regulated function of aerobic enzymes. However, under most conditions mitochondrial function is not the main factor limiting $\dot{V}_{O_2 \text{ MAX}}$ but inherited mitochondrial dysfunctions can be important.

1.6.3 Capillary density

The concept of increased capillary density relates to increases the overall surface area of the blood capillaries in working muscles. Capillary density increases with training and is correlated with an increase in $\dot{V}_{O_2 \text{ MAX}}$ (Andersen & Henriksson, 1977). The increase in capillary density makes it easier to improve cardiac output by reducing peripheral vascular resistance but also facilitates an increase in muscle transit time to improve capillary sarcolemmal diffusion (Saltin, 1985). Peripheral vascular disease and inactivity will reduce capillary density and may provide an additional mechanism to limit exercise capacity in such conditions.

It seems obvious therefore, by understanding the physiological mechanisms that limit $\dot{V}_{O_2 \text{ MAX}}$ it will be possible to trace a specific pathology when (CPET) is performed appropriately. However, the validity of the procedure can be compromised when tests are conducted without the relevant care and attention to potential artefacts that may influence the measurements being undertaken.

1.7 Measuring Oxygen Consumption

In the early twentieth century, $\dot{V}_{O_2 \text{ MAX}}$ measurements were relatively difficult and time consuming procedures. The $\dot{V}_{O_2 \text{ MAX}}$ was obtained by stressing the participants by high-intensity, intermittent, exercise to a point where sure the rate of oxygen consumption does not increase with increasing work load (Taylor *et al.*, 1955). The oxygen consumption was measured during steady-state gas collections over 30 seconds to one minute in mixing chambers known as Douglas bags (Douglas, 1911). This process required the participants to be quite determined since obtaining the steady-state response could take 2-3 minutes

before expired gases could be collected. Thus its use in patient populations was limited.

However, modern equipment facilitates the measurement of $\dot{V}_{O_2 \text{ MAX}}$ in diverse populations by analyzing the expired gas on a breath-by-breath basis. This approach allows the rise of oxygen consumption in proportion to increasing exercise intensity to be followed. These data gave better temporal resolution and demonstrated that during the initial seconds of effort that the relationship between oxygen consumption and energy expenditure is weak. It is anaerobic processes that make the greatest contribution to energy expenditure at this time (e.g. energy is produced from creatine phosphate).

Moreover, during the first 60-120 seconds of a constant workload increment the relationship between oxygen consumption and work load is non-linear. Firstly, there is an increase in pulmonary blood flow (phase I or the cardio-dynamic phase) before a rapid increase in oxygen consumption related to muscle extraction of O_2 (phase II or the oxygen uptake kinetic phase) before reaching steady-state (phase III) if the work load does not result in the accumulation of blood lactate. Therefore, even during breath-by-breath analysis, the correlation between oxygen consumption and work load only becomes linear during steady-state aerobic activities (Balady *et al.*, 2010). This has an important impact on the way (CPET) should be conducted.

A 'true' $\dot{V}_{O_2 \text{ MAX}}$ or \dot{V}_{O_2} plateau is usually unexpected in sedentary subjects and virtually absent in patient populations because of the high motivation levels needed for it to be observed (Midgley *et al.*, 2007). So the term $\dot{V}_{O_2 \text{ PEAK}}$ is often used when no \dot{V}_{O_2} plateau occurs with increasing work load (Taylor *et al.*, 1955) but the participants are unable to maintain the exercise effort (Lear *et al.*, 1999; Meyer *et al.*, 2005). It should be the term of choice when conducting symptom limited exercise tests in patient populations.

1.8 Other factors influencing oxygen consumption

As previously discussed, there are 'central' and 'peripheral' physiological factors that influence oxygen consumption and are amenable to interrogation for

establishing the aetiology of a patient's symptoms. However, other factors also influence oxygen consumption and may limit the relevance of data being generated during (CPET). These factors include:

1.8.1 Age

The highest $\dot{V}_{O_2 \text{ MAX}}$ measurement is generally made in the early twenties before gradually declining at about 10% per decade (Jackson *et al.*, 1995; Inbar *et al.*, 1994; Astrand *et al.*, 1997). Some studies report that the decline in maximum oxygen consumption is higher between 50 and 75 years, increasing to about 15% per decade (Shvartz & Reibold, 1990). However, by remaining physically active the age related decline in oxygen consumption is attenuated (Shvartz & Reibold, 1990; McGuire *et al.*, 2001). These age related effects reflect changes in maximal cardiac output following reduction in maximum heart rate and stroke volume (McGuire *et al.*, 2001).

1.8.2 Gender and Body Composition

Gender has a significant impact on $\dot{V}_{O_2 \text{ MAX}}$. Typically, women have a lower $\dot{V}_{O_2 \text{ MAX}}$ than men, generally by around 20-25% and this is not merely dependent on body weight. Women tend to have lower body mass and higher body fat so that part of their lower $\dot{V}_{O_2 \text{ MAX}}$ results from the oxidative capacity in a reduced muscle mass. However, other factors contribute too in as much as women have lower blood haemoglobin levels and small lung volumes that reduce the oxygen carrying capacity of the blood (Guenette *et al.*, 2007; Harms & Rosenkranz, 2008). Any significant change in body composition will alter the $\dot{V}_{O_2 \text{ MAX}}$ measurement and is not restricted to gender. Higher lean body mass within the male population will also tend to be associated with a higher $\dot{V}_{O_2 \text{ MAX}}$. Likewise, conditions associated with increased fat mass will have a lower $\dot{V}_{O_2 \text{ MAX}}$ partially attributable to the reduced proportion of muscle mass.

1.9 Heart rate (HR)

The correlation between heart rate and oxygen consumption is fairly linear between 50-90% of maximum heart rate. However, at the onset of exercise stroke volume increases due to the increased venous return from the muscle pump. Consequently, the early heart rate response is blunted and the cardiac output response is driven by the change in stroke volume. Furthermore, the heart rate response will flatten just before the maximum level to facilitate ventricular filling (Astrand, 1976).

A good indicator that a test participant is at maximal effort is his or her heart rate reserve. Heart rate reserve being the difference between predicted maximum heart rate and heart rate at peak exercise. Heart rate maximum HR_{max} is commonly predicted from the participants age ($220 - \text{age}$) but there is considerable variability between participants and maximal effort is often accepted when participants achieve 85-95% of age-predicted maximum (Lear *et al.*, 1999; Gibbons *et al.*, 2002). Heart rate reserve is usually found to be less than 15 beat/min in the healthy (Franklin *et al.*, 2000; Fletcher *et al.*, 2001). However, in some clinical conditions the heart rate reserve can be increased so that a reduction in maximum heart rate may contribute to reduced $\dot{V}_{O_2 \text{ MAX}}$. Perhaps the most common example would be with the use of β -blockers in the treatment of coronary heart disease.

1.10 Pulmonary ventilation (\dot{V}_E)

Pulmonary ventilation \dot{V}_E is the product of breathing frequency B_f in a minute and tidal volume V_t . Pulmonary ventilation can increase from 7 l/min at rest to over 100 l/min at maximum effort in health. Increasing both B_f and V_t contribute to increasing pulmonary ventilation at low exercise intensities (Gallagher *et al.*, 1987) by and up until 70-80% of maximum (Gallagher *et al.*, 1987; Johnson *et al.*, 1992) and beyond this point there is a disproportionate rise in B_f (ATS/ACCP, 2003).

This means that \dot{V}_E correlates linearly with work load and \dot{V}_{O_2} until there is evidence of blood lactate accumulation. After this point there is an increase in

\dot{V}_E that is higher than the corresponding increase in \dot{V}_{O_2} (Mezzani *et al.*, 2009). At maximum effort during (CPET) the measured \dot{V}_E is usually around 70% of the estimated maximum voluntary ventilation (MVV) in health (ATS/ACCP, 2003) and therefore a breathing reserve of around 10-15 l is retained.

In clinical conditions *e.g.* chronic obstructive pulmonary disease COPD, the breathing reserve ($MVV - \dot{V}_{E\text{ PEAK}}$) can be reduced so that the respiratory effort limits the patients ability to achieve $\dot{V}_{O_2\text{ MAX}}$. The ventilatory limit to exercise is not restricted to maximum effort and dividing \dot{V}_E by \dot{V}_{O_2} represents a common measure of ventilatory efficiency. In health, $\dot{V}_E / \dot{V}_{O_2}$ is around 25-30 at rest, reduces during sub-maximal exercise and rises to around 35 at maximal effort (ATS/ACCP, 2003).

1.11 Carbon dioxide production (\dot{V}_{CO_2})

One of the main functions in the respiratory system is to remove carbon dioxide CO_2 from the body. Obviously, the increase in CO_2 partial pressure $PaCO_2$ is the main factor that enhances pulmonary ventilation through the stimulation of peripheral chemoreceptors (ATS/ACCP, 2003). The production of CO_2 is the result of cellular respiration and as such is correlated with exercise intensity (Astrand & Rodahl, 1986). Early in graded exercise effort CO_2 production increases linearly with oxygen uptake, however, with increasing effort the anaerobic component to energy metabolism will mean that CO_2 production increases in relation to O_2 uptake.

In addition, the relationship between \dot{V}_{CO_2} and \dot{V}_E is linear with the slope of the $\dot{V}_{CO_2} / \dot{V}_E$ relationship measured at 24.6 ± 2.4 in healthy subjects during aerobic metabolism (Wasserman K, 1999). During exercise with anaerobic metabolism, the excess CO_2 production stimulates ventilation to buffer CO_2 and consequently the slope of the $\dot{V}_{CO_2} / \dot{V}_E$ relationship becomes steeper and has been measured at 35.5 ± 3.6 in healthy subjects (Gademan *et al.*, 2008). However, Sun *et al* have observed slopes <30 in normal volunteers without considering age and sex differences (Sun *et al.*, 2002). Whatever the magnitude of the change in the $\dot{V}_{CO_2} / \dot{V}_E$ relationship, its presence is important and a steepening of this relationship has been termed the respiratory compensation point RCP and used

as an objective measure to indicate the onset of anaerobic exercise (Wasserman *et al.*, 1977).

In clinical populations, e.g. in coronary artery disease (CAD), peripheral vascular disease (PVD), obesity and chronic obstructive pulmonary disease (COPD) a very steep $\dot{V}_{\text{CO}_2}/\dot{V}_{\text{E}}$ relationship (>60) can indicate limitations in aerobic capacity (Yasunobu *et al.*, 2005).

1.12 Respiratory exchange ratio (RER)

The ratio of \dot{V}_{CO_2} and \dot{V}_{O_2} is called the respiratory exchange ratio RER. This ratio is similar to the respiratory quotient RQ during steady state aerobic exercise where the CO_2 production is equal to or lower than the O_2 uptake at the mouth. Since it is technically difficult to determine RQ, many exercise physiologists and clinicians use RER to estimate non-protein substrate utilisation during aerobic exercise. An RER value close to 1.0 represents predominantly carbohydrate fuelled metabolism, an RER value close to 0.7 represents predominantly fat fuelled metabolism and between these values represents mixed substrate metabolism (ATS/ACCP, 2003; Knight *et al.*, 1995). However, during anaerobic metabolism the RER is in excess of 1.0 due to no metabolic production of CO_2 and so no inference can be taken in relation to substrate utilisation. Nevertheless, the ratio is useful in establishing relative exercise intensity and maximal efforts are associated with RER values in excess of 1.15 (Issekutz, 1962).

1.13 Exercise Modality

Exercise can take many forms and includes walking, running, cycling, swimming, skiing and rowing. However, despite the wide range of exercise modalities available for (CPET) the choice in clinical practice is generally restricted to the use of treadmills and cycle ergometers. Both modalities have advantages and disadvantages and as a result, the choice of exercise modality tends to be based on cultural biases.

1.13.1 Treadmill testing

Using a treadmill for (CPET) has the advantage that it simulates relatively natural activities of walking and running. The use of a relatively large muscle mass during this sort of exercise tends to lend itself to objective measures of whole body O₂ uptake that reflects normal every day activities and allows true maximal effort and higher measured values. The participant's body weight needs to be supported during the exercise test and so obese patients are disadvantaged reflecting their daily activities. Treadmills are the most commonly exercise modality in North America and the United Kingdom where recreational cycling is, perhaps, less popular than in mainland Europe (Hermansen & Saltin, 1969;Stuart Jr & Ellestad, 1980;Taylor *et al.*, 1955).

On other hand, treadmills tend to be more expensive, require more space, are difficult to move and are perhaps less secure for participants with functional limitations (ATS/ACCP, 2003). Moreover, it can be difficult to determine accurate work loads during treadmill studies (and, therefore, the metabolic cost) (ATS/ACCP, 2003). Typically work load is simple expressed in as speed (in km/h or miles/h) and incline (in % gradient), however, the American College of Sports Medicine do provide mathematical equations to calculate estimated work load on the treadmill (Williams and & Wilkins, 1995).

1.13.2 Cycle Ergometer Testing

Using a cycle ergometer for CPET has some advantages. It is often preferred in clinical exercise laboratories where frail or infirm patients are unable to walk safely on a treadmill (Lear *et al.*, 1999). Cycle ergometers are lighter and easier to move than treadmills, they are less costly and take up less space. In addition, by being seated during exercise there is less motion in the upper body which makes blood sampling and blood pressure recording easier. Furthermore, the calculation of work load during (CPET) is fairly accurate (Gibbons *et al.*, 2002;Lear *et al.*, 1999;Northridge *et al.*, 1990)

However, cycling is weight independent and does not so accurately reflect normal day-to-day activities. Furthermore, power output and whole body O₂

uptake will reflect body composition (mostly lean body mass) and can lead to significant variability in the measured $\dot{V}_{O_2 \text{ PEAK}}$

(Northridge *et al.*, 1990). This can mean that lower limb weakness and/or poor cycling technique may mean that some participants are limited during cycle ergometer by the exercise modality and do not achieve a true maximal effort (Gibbons *et al.*, 2002). Even in participants that are accustomed to cycling it is found that treadmill tests tend to provide higher measured $\dot{V}_{O_2 \text{ PEAK}}$ (by 10-20%) and HR_{max} (5-20%) values than in cycle ergometer studies (Whipp *et al.*, 1981;ATS/ACCP, 2003;Buchfuhrer *et al.*, 1983;Hermansen & Saltin, 1969).

1.13.3 Exercise protocols

In addition to the exercise modality influencing the physiological response being measured during (CPET), the precise nature of the exercise protocol can influence the measurements obtained. The exercise protocol dictates the time between stages of exercise and the pattern of work load increment during the exercise. Previous studies have shown stage duration and the pattern of work load increment during (CPET) can influence the values of power output and O_2 uptake measured during and at the cessation of the exercise tests (Bishop *et al.*, 1998;Roffey *et al.*, 2007).

In designing a protocol to determine $\dot{V}_{O_2 \text{ PEAK}}$ during (CPET), one of the main aims would be to obtain work load at which \dot{V}_{O_2} plateaus despite increasing work load. This could involve continuous or discontinuous protocols with stage durations lasting seconds (as in ramp protocols) or minutes. It has been established that altering the stage duration will produce the same $\dot{V}_{O_2 \text{ PEAK}}$ in healthy volunteers despite big differences in total exercise duration (from 10-30 minutes) (Midgley *et al.*, 2007).

However, the inappropriate selection of an exercise protocol might lead to over or under estimation of exercise capacity (Maeder *et al.*, 2006). In one study, $\dot{V}_{O_2 \text{ PEAK}}$ and the anaerobic threshold was measured in healthy untrained and highly trained volunteers using three different exercise protocols (the Astrand, Bruce and Costill/Fox protocols). The study showed a wide range of test durations from

the exercise protocols in both groups. In the untrained group, the Costill/Fox protocol duration was only 4.9 ± 0.3 minutes compared to 9.8 ± 0.5 and 12.4 ± 0.4 in the Astrand and Bruce protocols respectively. As might be expected, the exercise durations of the trained group were longer but again showed a similar range in test duration, with only 10.4 ± 0.4 minutes for the Costill/Fox protocol compared to 14.5 ± 0.5 and 17 ± 0.5 in the Astrand and Bruce protocols respectively. The $\dot{V}_{O_2 \text{ PEAK}}$ was measured the same in the untrained group independent of the protocol whereas the trained group had a lower $\dot{V}_{O_2 \text{ PEAK}}$

measured during the Bruce protocol. Moreover, the anaerobic threshold was measured as being lower in both groups during the Bruce protocol when compared to the others (Kang et al., 2001).

Others have also shown that the pattern of work load increment can influence the $\dot{V}_{O_2 \text{ PEAK}}$ measured during different exercise protocols. Large increments in work load tend to cause participants to end the test early because of muscle fatigue in the legs or thighs. Smaller work load increments allow the participants to achieve higher peak work rates and therefore higher $\dot{V}_{O_2 \text{ PEAK}}$ (Buchfuhrer *et al.*, 1983). Clinical observations also suggest that increasing work rate too rapidly can lead to an over-prediction of functional capacity and can negatively impact on the sensitivity of an exercise test to detect coronary artery disease (Fletcher *et al.*, 2001; Myers *et al.*, 1991). These observations have meant that ramp exercise protocols which facilitate low work load increments may be more useful in patient populations (Myers & Bellin, 2000; Myers *et al.*, 1991).

For that reason, ramp exercise protocols (*i.e.* with a continuous low increment work load increase) are commonly utilised in research laboratories conducting (CPET) (Blackie *et al.*, 1991). The functional capacity is more accurately estimated using ramp protocols (Myers & Bellin, 2000; Maeder *et al.*, 2006) and the relationship between work rate, oxygen uptake and other physiological responses are better correlated than with more traditional exercise protocols such as the Bruce (Bruce, 1971) or Balke protocols (Balke & Ware, 1959).

There have been many trials that aimed to design standardised exercise protocols for both treadmill (Porszasz *et al.*, 2003; Hunt, 2008; Wolthuis *et al.*,

1977) and cycle ergometer (Stockhausen *et al.*, 1997) testing. With protocols developed for use over a wide range of age categories (Benzo *et al.*, 2007; Northridge *et al.*, 1990; Buchfuhrer *et al.*, 1983; Northridge *et al.*, 1990), functional capacity (Buchfuhrer *et al.*, 1983; Northridge *et al.*, 1990) and clinical status (Benzo *et al.*, 2007; Wolthuis *et al.*, 1977).

However, treadmill ramp protocols are a controversial issue due to the difficulty in their implementation across the mainstream clinical laboratories. Typically, a graded incremental treadmill protocol will have relatively infrequent changes in treadmill speed and/or gradient and can be catered for by a relatively simple treadmill (Franklin *et al.*, 1983). Whereas, combining curvilinear increments in treadmill gradient with linear increases in speed that might be suitable for application in patient populations is often beyond the mechanical capabilities of common clinical treadmills (Porszasz *et al.*, 2003). Moreover, the equations used to calculate an age-predicted ramp work load may not be applicable across the range of clinical population. However, other studies have reported exponential increments in workload can be applied on simple treadmill and cycle ergometers to obtain a constant workload increment that provides reproducible sub-maximal and maximal data in wide range of patient populations (Northridge *et al.*, 1990)

What should not be overlooked though is the fact that, in clinical settings, it is not unusual for symptom limited exercise testing to be undertaken in patient populations without respiratory gas analysis. In such cases, even though different exercise protocols using different stage duration could elicit the same $\dot{V}_{O_2 \text{ PEAK}}$ the lack of a consistent exercise duration will prevent clinicians from assessing functional capacity or monitoring the response to a drug therapy (Myers & Bellin, 2000). Consequently, a standardised approach to clinical testing protocols has developed and the vast majority (>80%) of treadmill tests use the Bruce or Modified Bruce protocol in (CPET) (Myers & Bellin, 2000; Bruce, 1971; Will & Walter, 1999). The widespread use of these protocols have allowed significant accumulation of normative data during (CPET) and can allow an abnormal treadmill exercise response to be established in coronary artery patients without respiratory gas analysis (Weiner *et al.*, 1987). This is despite the fact that the estimation of exercise capacity using the Bruce protocol was found to be less accurate with cardiac patients (Myers *et al.*, 1991).

1.14 Exercise Duration

Total exercise duration is an essential feature in the design of an exercise protocol; typically prolonged exercise durations are avoided to reduce the risk of boredom within the test participants or even the (CPET) supervisor.

Additionally, it has been suggested that when (CPET) extends to more than 17 minutes then a low $\dot{V}_{O_2 \text{ PEAK}}$ is recorded due to increased body temperature, dehydration, intercostal muscle fatigue and generalised discomfort (Buchfuhrer *et al.*, 1983). Buchfuhrer *et al* also suggest that short test durations (less than 8 minutes) can result in low recorded $\dot{V}_{O_2 \text{ PEAK}}$ measurements that might be due to lower limb muscle soreness. Therefore, the authors suggested that exercise durations between 8 and 17 minutes are the optimum to obtain $\dot{V}_{O_2 \text{ PEAK}}$ measurements and this study forms the basis for current ACSM guidelines on clinical exercise testing.

In contrast to this study, there is evidence that short duration exercise of less than 60 seconds can elicit relatively high $\dot{V}_{O_2 \text{ PEAK}}$ measurements (McLellan, 1985; Froelicher, Jr. *et al.*, 1974). Whereas, Roffey *et al* showed a decrease in measured $\dot{V}_{O_2 \text{ PEAK}}$ when total exercise duration was more than 10 minutes (Roffey *et al.*, 2007). The situation being further complicated by the work of Fairshter *et al* that showed no significant difference in $\dot{V}_{O_2 \text{ PEAK}}$, \dot{V}_E and anaerobic threshold when using protocols with different exercise durations (Fairshter *et al.*, 1983).

In the clinic, the minimum acceptable duration that will provide useful exercise data for frail patients is suggested to be three minutes (Redwood *et al.*, 1971). However, despite controversy in the literature, the current ACSM guidelines suggest that (CPET) should last between eight and twelve minutes to elicit a true maximal performance. There are no stated guidelines on the performance of sub-maximal tests.

1.15 Sub-maximal markers of exercise performance

The performance of a maximal exercise effort can be undesirable or even impossible in certain medical conditions *e.g.* in patients with musculo-skeletal disorders, where they have neurological disorders, where they are excessively

de-conditioned or are contra-indicated to maximal efforts due to cardiopulmonary limitations (Esc A working Group, 1993; Franklin *et al.*, 2000). In such situations, sub-maximal (CPET) may be appropriate to assess the cardiopulmonary limitation to exercise (Lear *et al.*, 1999). The most common sub-maximal markers employed in exercise physiology are ventilation at anaerobic threshold VAT, breathing reserve index at anaerobic threshold (BRI_{AT}) and the oxygen uptake efficiency slope (OUES).

1.15.1 Ventilation at Anaerobic Threshold (VAT)

Determining the anaerobic threshold has become a routine physiological measurement during (CPET) to assess functional capacity in sporting, healthy and patient populations (Davis *et al.*, 1976; Davis *et al.*, 1979; Davis, 1985). It is now a criterion measure used to assess the level of impairment/disability for work, since the ability to maintain high metabolic rate before the onset of anaerobic acidosis predicts enough cardiopulmonary reserve to work for at least 8 hours (Hansen *et al.*, 1984; ATS/ACCP, 2003).

At the beginning of 1970s, the use of respiratory gas analysis was developed to provide a non-invasive technique to assess relative exercise intensities (Wasserman *et al.*, 1973). The anaerobic threshold (AT) was defined as the level of $\dot{V}O_2$ during (CPET) exercise testing at which an increased production of lactic acid could be identified. Davis *et al.* demonstrated that specific changes in expired gas concentrations correlated well with the increase in blood lactate levels observed during progressive cycle ergometer exercise. The increase in blood lactate concentration is termed the lactate threshold (LT) and represents the switch from aerobic to anaerobic energy production during progressive exercise (Davis *et al.*, 1976). As exercise intensity increases, the relative proportion of energy production through anaerobic mechanisms increases and lactate accumulates in the blood and muscles tissue (Wasserman *et al.*, 1973; Wasserman *et al.*, 1985).

Many studies have since reported that the (LT) is strongly correlated with ventilatory anaerobic threshold VAT (Aunola & Rusko, 1986; Davis *et al.*, 1976; Yoshida *et al.*, 1981). The term VAT actually refers to the onset of exercise induced hyperventilation during (CPET). This increase in \dot{V}_E is a

homeostatic response to deal with the consequences of the excess lactate production which can dissociate to release H^+ ions from lactic acid into the blood stream. The H^+ ions are buffered by bicarbonate and release CO_2 (Wasserman & Mcilroy, 1964). This buffering of lactic acid results in extra CO_2 production over that produced by aerobic metabolism and increases the arterial CO_2 partial pressure $PaCO_2$. The increase in $PaCO_2$ stimulates excess ventilation that follows on from the lactate threshold (Wasserman *et al.*, 1975).

Different gas analysis techniques have been suggested to identify the VAT. Previous studies have identified non-linear $\dot{V}CO_2$ output at the lungs and an increase in RER quickly following the (LT) (Issekutz & Rodahl, 1961; Issekutz, 1962). However, it is difficult to use these markers to determine the start of metabolic acidosis because during breath-by-breath analysis it is hard to identify the exact point at which \dot{V}_E , \dot{V}_{CO_2} and RER increase (Wasserman *et al.*, 1973; Davis *et al.*, 1976).

In research laboratories, identification of the VAT is often considered from two view points during (CPET); using the ventilatory equivalent for \dot{V}_{O_2} ($\dot{V}_E / \dot{V}_{O_2}$) and end-tidal PO_2 ($P_{ET}O_2$) to monitor relative exercise intensity. Essentially, these two variables decrease during the initial stages of exercise due to a decrease in the ratio of physiological dead space to tidal volume (V_D/V_T). Subsequently, both $\dot{V}_E / \dot{V}_{O_2}$ and $P_{ET}O_2$ begin to increase in proportion above VAT (Wasserman & Whipp, 1975; Yoshida *et al.*, 1981).

In contrast, the ventilatory equivalent for \dot{V}_{CO_2} ($\dot{V}_E / \dot{V}_{CO_2}$) and end-tidal PCO_2 $P_{ET}CO_2$ remains constant through a period of isocapnic buffering. Isocapnic buffering is when there is a lack of ventilatory compensation to a developing metabolic acidosis and is the result of bicarbonate buffering of the lactic acidosis. Thus an increase in $\dot{V}_E / \dot{V}_{O_2}$ without a corresponding increase in $\dot{V}_E / \dot{V}_{CO_2}$ gives a clear indication that the (LT) has been passed (Caiozzo *et al.*, 1982; Yoshida *et al.*, 1981; Wasserman & Whipp, 1975).

In clinical laboratories, however, simpler approaches have been adopted that allow automated algorithms within gas analysis systems to indicate the VAT. The most common applied is the V-slope method (Beaver *et al.*, 1986). A V-slope can

be obtained by plotting the relationship between \dot{V}_{CO_2} and \dot{V}_{O_2} . The relationship between \dot{V}_{CO_2} and \dot{V}_{O_2} is linear below VAT but becomes steeper as \dot{V}_{CO_2} increases to buffer the metabolic acidosis. It is the break point in this linear relationship that is used to define the VAT point.

Despite the various ways of defining VAT from respiratory gas analysis, there is a fairly consistent view on its benefit for determining functional capacity. In general, VAT occurs between 50-65% of $\dot{V}_{\text{O}_2 \text{ PEAK}}$ (Wasserman *et al.*, 1973; Davis *et al.*, 1979) with 40% of $\dot{V}_{\text{O}_2 \text{ PEAK}}$ suggested to be the lower end of the VAT range in healthy sedentary individuals (Sue & Hansen, 1984). In clinical situations, VAT is a poor prognostic indicator when measured at a \dot{V}_{O_2} of less than $11 \text{ ml}^{-1} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ (Sinclair *et al.*, 2009) and can provide a better measure of functional capacity than $\dot{V}_{\text{O}_2 \text{ PEAK}}$.

As $\dot{V}_{\text{O}_2 \text{ PEAK}}$ reduces with age it is unsurprising that the absolute value (in l/min) of \dot{V}_{O_2} at VAT is reduced with age (Jones *et al.*, 1985). However, the decline in $\dot{V}_{\text{O}_2 \text{ PEAK}}$ with age is greater than that of VAT so that the relative value (in %) increases slightly with age (Astrand & Rodahl, 1986). Also, the determination of VAT in relative terms is not influenced by exercise modality or gender (Buchfuhrer *et al.*, 1983; Bhambhani & Maikala, 2000; Habedank *et al.*, 1998).

Importantly, the VAT may be influenced by the design of the exercise protocol employed with low values being measured a small increment ramp test ($8 \text{ W} \cdot \text{min}^{-1}$) compared to a larger increment ramp test ($65 \text{ W} \cdot \text{min}^{-1}$) (Hughson & Green, 1982). Whereas, other studies suggest similar results with one and four minute time intervals (Wasserman *et al.*, 1973; Yoshida, 1984).

1.15.2 Oxygen Uptake Efficiency Slope (OUES)

The oxygen uptake efficiency slope (OUES) was originally described by Baba *et al.* for conducting (CPET) in paediatric patients that were unmotivated to perform maximal testing procedures (Baba *et al.*, 1996). The OUES reflects the relationship between \dot{V}_{O_2} and \dot{V}_{E} during incremental exercise and is best described by a single exponential function where the exponent gives the OUES. The transformed logarithmic regression is linear in almost all subjects, and,

therefore, the OUES (unlike $\dot{V}_{O_2 \text{ MAX}}$) does not require a true maximal effort for its valid estimation. The OUES is not significantly different between maximal effort and 75% or 90% of total exercise duration and is strongly correlated with $\dot{V}_{O_2 \text{ MAX}}$ in patients who can exert maximal and sub-maximal efforts (Hollenberg & Tager, 2000).

The major factors that influence the OUES are CO_2 production (derived from muscle aerobic metabolism as well as from the pH buffering function of bicarbonate), arterial pCO_2 (CO_2 setpoint) and physiologic pulmonary dead space ventilation. Untrained volunteers or patients with medical conditions that result in early lactic acidosis during exercise will have a diminished OUES, as will those with poor lung structural integrity or inadequate pulmonary perfusion.

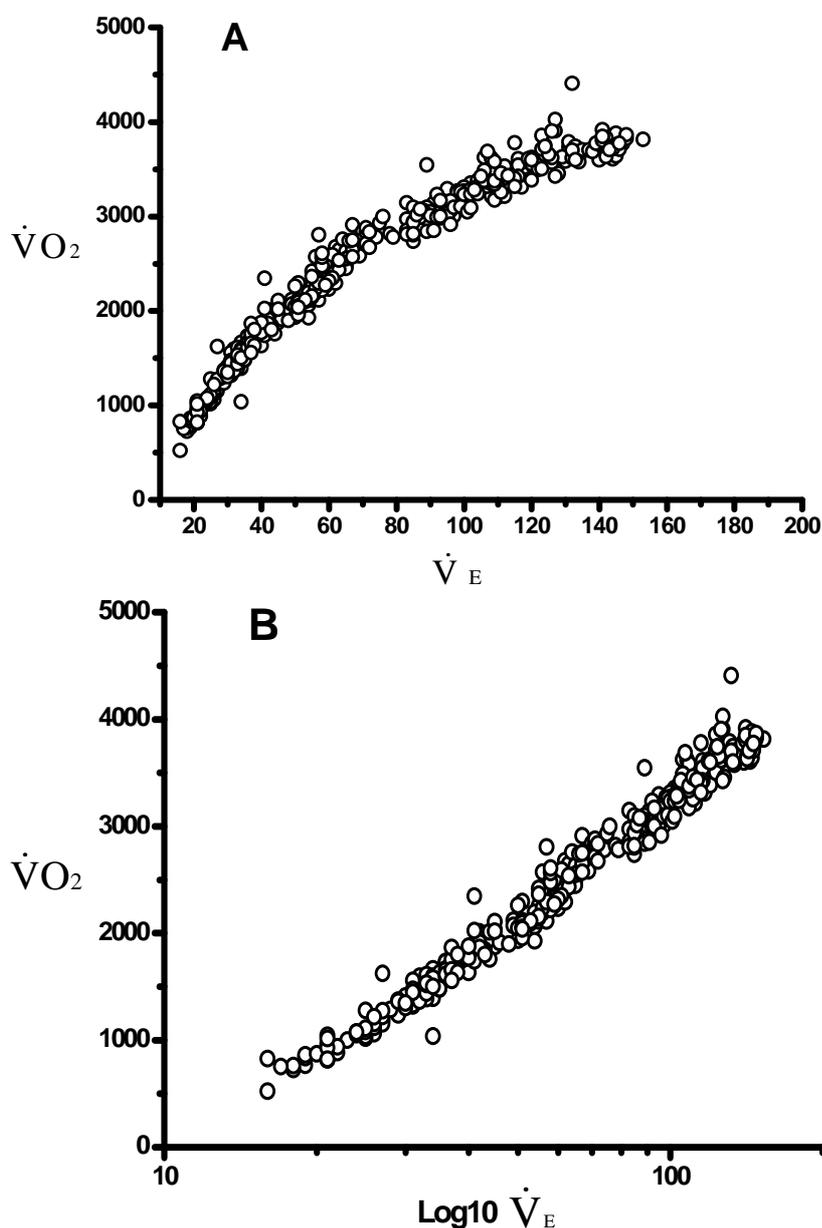


Figure 1-1 Illustrates the slope before (A) and after (B) log plot of \dot{V}_E l.min⁻¹ against $\dot{V}O_2$ ml⁻¹.kg⁻¹.min⁻¹.

1.15.3 Breathing Reserve Index(BRI)

A further novel marker of functional capacity is the breathing reserve index (BRI). BRI is the fraction of maximal voluntary ventilation (MVV) used at peak exercise \dot{V}_E and is known as the peak breathing reserve index (BRI_{peak}).

Alternatively, the fraction of (MVV) used at the anaerobic threshold has been termed the breathing reserve index at anaerobic threshold (BRI_{AT}) (Tantisira *et al.*, 2002; Sexauer *et al.*, 2003; Ross, 2003; Medoff *et al.*, 1998).

The study by Medoff *et al* demonstrates that (BRI_{AT}) is sensitive in identifying ventilatory limitations to exercise in a range of patient populations. Specifically, they found that (BRI_{AT}) could distinguish between pulmonary mechanical limitations to exercise in chronic obstructive pulmonary disease (COPD) and that caused by cardiovascular limitations. Furthermore, Sexauer *et al* found that a (BRI_{AT}) of 0.29 could discriminate between cystic fibrosis patients whose exercise capacity was limited by ventilation or non-ventilation (Sexauer *et al.*, 2003). Whereas, a (BRI_{AT}) of 0.42 discriminates between COPD patients, cardiac patients and healthy volunteers (Medoff *et al.*, 1998).

Measuring (MVV) accurately is a fundamental factor that influences the accurate determination of (BRI_{peak}) and (BRI_{AT}) (Sexauer *et al.*, 2003) but it can be difficult accurately measure and requires good co-operation from patients (Kor *et al.*, 2004). Consequently, most clinicians will estimate (MVV) using a range of formulae e.g. $FEV_{1 \times 35}$ (Gandevia & Hugh-Jones, 1957), or $FEV_{1 \times 41}$ (Miller *et al.*, 1959) or $FEV_{1 \times 45.12-15.85}$ (Kor *et al.*, 2004), where FEV_1 represents the forced expiratory volume in one second.

1.16 The rationale for the current programme of work

The current recommendation from the ACSM for the conduct of (CPET) suggests that exercise duration lasts between 8-12 minutes (Buchfuhrer *et al.*, 1983;ATS/ACCP, 2003;Davis *et al.*, 1982). These recommendations are based on data from a single study (Buchfuhrer *et al.*, 1983) with limited sample size and subjective analysis focused on maximal aerobic performance. In this study, twelve men completed various test protocols (using one minute stage increments) on both treadmill and cycle ergometers until they reached volitional exhaustion. Breath by breath data was analysed but only reported for five relatively young healthy, male subjects (36 ± 9.7 years old). They performed five treadmill and three cycle ergometer tests and the highest $\dot{V}_{O_2 \text{ PEAK}}$ was recorded with a work rate increment that brought the subjects to the limit of their tolerance within 10 ± 2 minutes. These data suggested that the highest $\dot{V}_{O_2 \text{ MAX}}$ values were achieved between 8-17 minutes. This analysis may be biased.

More recent studies have suggested no significant effect on the $\dot{V}_{O_2 \text{ MAX}}$ obtained using exercise durations out with the current guidelines (Lepretre *et al.*, 2004). Moreover, it is obvious from the preceding discussion that not all clinical (CPET) procedures should have the aim to obtain maximal effort and that relatively little is published in relation to the influence of exercise duration *per se* on markers of functional capacity. The aim of this thesis is, therefore, to add to the data available on the effects of exercise duration on the measurement of $\dot{V}_{O_2 \text{ PEAK}}$ from the previous study (Buchfuhrer *et al.*, 1983). Moreover, the data will include a greater range of age, gender and functional capacity than is currently available to make them more relevant to clinical application.

Furthermore, the effects of exercise duration will be investigated on common sub-maximal markers of functional capacity to determine whether new ACSM guidelines should be established to specifically address sub-maximal testing procedures.

1.17 Hypothesis

The working hypothesis is that exercise duration *per se* does not influence the physiological response to exercise so that maximal and sub-maximal markers of exercise performance should not change. However, reducing exercise duration may limit the accuracy and reliability of measuring ventilatory threshold, oxygen uptake efficiency slopes and breathing reserve index. These are commonly used in clinical practice as sub-maximal indices of exercise capacity.

CHAPTER TWO

Methods

2 Methods

The strategy for developing the treadmill and cycle ergometer protocols was developed from a pilot study using four participants.

2.1 Participants and methods for pilot study:

Four sedentary males participated in this pilot study. They were all free of known cardiac disease, cerebrovascular disease and musculoskeletal impairment. None was physically active or engaged in any form of regular exercise or at high risk from exercise testing according to the guideline of ACC/AHA Guidelines for exercise testing (Franklin *et al.*, 2000; Gibbons *et al.*, 2002).

2.1.1 Design of pilot study:

Three of the participants completed at least six continuous breath-by-breath respiratory gas measurements studies using a metabolic cart (Health care - Oxycon Pro). They completed three tests on a treadmill (Woodway GmbH D 79576) using a modified Bruce protocol (with simultaneous changes in speed and gradient) (Porszasz *et al.*, 2003). The duration of the individual work rate stages were altered (at 7s, 8s, 13s, 15s and 17s intervals) to determine appropriate stage durations for the targeted total exercise duration. A similar rationale was applied to the development of a cycle ergometer protocol where the duration of the workload increment was kept constant (at 60s) and the magnitude of the workload increment was increased (from 10W.min⁻¹ to 30W.min⁻¹).

2.2 Subjects

Study participants were recruited between July 2009 and May 2010 through advertisements in University of Glasgow website (My Glasgow) and local media. They volunteered from University of Glasgow staff and students and from the general population of Glasgow city. The volunteers were multi-cultural and comprised a wide range of different nationalities including Britain, Saudi Arabia, Malaysia, Denmark, Iran, Libya, Indonesia, Pakistan, Spain and Nigeria.

The volunteers were healthy male and females aged between 21 and 57 years. None of the volunteers participated in competitive athletics or engaged in any form of regular exercise. They were all free from cardiovascular, respiratory and musculo-skeletal disease or any other medical disorder that might prevent them from achieving a maximum exercise performance.

All participants provided informed consent (appendix 1) and were fully informed about the purpose of the study (appendix 2). A standard physical activity form was given to the subjects. This form commonly used in exercise physiology department at Glasgow university. They completed questionnaires prior to (CPET) that provided information on their personal details, physical characteristics and recorded their current levels of physical activity e.g. their estimated intensity of daily exercise activity and the number of exercise sessions that they engage in per week (appendix 3).

In addition, the questionnaires recorded their medical history to help identify any medical condition that might be a contraindication to (CPET). The study was approved by Ethics Committee for Non Clinical Research Involving Human Subjects materials or data (appendix 4). Any subject suffering from an acute or chronic medical condition was excluded from the study.

Extra medical investigations were carried out for both male and female volunteers over 35 years. Their resting blood pressure and electrocardiograph (ECG) was recorded and they underwent a clinical examination by a physician before proceeding to (CPET).

2.3 Physical characteristic of all subjects recruited to the study

In total 19 males and 12 females volunteered to participate in the current study and their descriptive characteristics are presented in (table 2.1). In total, male volunteers performed 76 ramp exercise protocols (38 on a treadmill and 38 on a cycle ergometer) and female volunteers performed 60 ramp exercise protocols (31 on a treadmill and 29 on a cycle ergometer). These statistics illustrate that not all volunteers completed the same number of (CPET)s during the study.

On recruitment, the volunteers were asked to complete three treadmill tests and three cycle ergometer tests with the three tests on each exercise modality being completed within a two week period. Six male and two female volunteers did not complete all three treadmill tests scheduled for the study and five male and two female volunteers did not complete all three cycle ergometer scheduled tests. The data from these subjects were included in some portions of the subsequent analysis and consequently the subject characteristics of specific groups being analysed will be presented as appropriate. However, the results from one male and one female volunteer were excluded from all analysis due to their tests being completed outside the two weeks limit.

Variable	Male	Female
Number	19	12
Age (years)	30.5±6.3 (21-42)	36±10.4 (24-57)
Weight (kg)	80.0±10.2 (60-103)	61.0±8.0 (48-77)
Height (cm)	176.0±5.7 (163-188)	163±7.8 (145-175)
Treadmill $\dot{V}_{O_2 \text{ PEAK}}$ (l.min ⁻¹)	3.48±0.56 (2.38-4.54)	2.41±0.61 (1.48-3.63)
Treadmill $\dot{V}_{O_2 \text{ PEAK}}$ (ml ⁻¹ .kg ⁻¹ .min ⁻¹)	46.1± 6.1 (31-59)	39.7±8.6 (25-56)
Cycle ergometer $\dot{V}_{O_2 \text{ PEAK}}$ (l.min ⁻¹)	3.00±0.53 (2.00-3.90)	2.02±0.56 (1.31-3.40)
Cycle ergometer $\dot{V}_{O_2 \text{ PEAK}}$ (ml ⁻¹ .kg ⁻¹ .min ⁻¹)	38.2±6.6 (26-51)	33.0±8.1 (22-52)
BSA* (m ²)	1.97±0.14 (1.65- 2.22)	1.66±0.12 (1.47-1.87)
BMI [#] (kg. m ²)	25.8±3.3 (21-34)	23±3.4 (18-29)

Table 2.1 Physical characteristics of all study volunteers. Data are presented as mean±SD (where appropriate) and the range is given in parenthesis. * BSA is Body Surface Area according to Mosteller formula. # BMI is Body Mass Index.

2.4 Experimental design

After the volunteers had successfully completed the initial health screening procedures they were scheduled for at least eight different (CPET) sessions. They were scheduled for one familiarisation session on each exercise modality and three experimental (CPET) sessions on each exercise modality. The order of

ramp exercise test for the studies were randomised to avoid order effects resulting from practice, learning or boredom in the participants (Gaito, 1961).

Each experimental visit was at least 48 hours apart to avoid the effects fatigue or muscle soreness/discomfort influencing the subsequent testing session. However, all three tests on one exercise modality were completed within a two weeks period to ensure no change in the volunteer's fitness. On testing days, volunteers were instructed not to exercise in the preceding 24 hours, not to smoke, to avoid alcohol and caffeine in the preceding 24 hours and to avoid a heavy meal in the four hours before the test.

Every (CPET) protocol employed consisted of four stages; resting, unloaded, test ramp and recovery. In both exercise modalities, (CPET) was preceded by 3-4 minutes of resting measurements to ensure that the volunteer has become accustomed to the mouthpiece and nose clip before starting to exercise. To ensure this, both RER and \dot{V}_E was monitored during the resting stage and the unloaded stage was not initiated until the RER was in the range 0.7-0.89 and \dot{V}_E was between 10 and 15 l.min⁻¹. The importance of an unloaded stage (a steady state, low metabolic requirement, initial exercise period) when designing a ramp exercise protocol is to avoid hyperventilation at the onset of the ramp exercise stage which facilitates a more accurate determination of the VAT break point (Ozcelik *et al.*, 1999).

After the unloaded stage, the volunteers were encouraged to reach their maximal capacity for each of the test ramp stages employed. A standard command was used for all participants (*i.e.* Do as much as you can!) was used and this encouragement was initiated after the VAT break point. The importance of using identical commands to encourage maximum effort was to avoid any bias in eliciting a maximal response. The volunteers were then encouraged at regular intervals to continue exercising until they were unable to maintain the progressively increasing of work rate. The inability to maintain the imposed work rate was indentified as a drop in cadence below 50 revs per minute on the cycle ergometer or the inability to maintain position towards the front of the treadmill. When the participant had reached their maximum exercise capacity, the test ramp protocol was terminated and an immediate active recovery stage

initiated and maintained for five minutes. Details of the work loads in each stage are given later in this chapter.

All (CPET) was performed in a comfortable air conditioned laboratory environment with a mean temperature of 21°C and a range of 18- 23°C and the following measurements were carried out before conducting the first test ramp protocol.

2.5 Body mass and height

On entering the laboratory, the volunteer's body weight was measured, while wearing light underwear, to the nearest 0.1 kg using calibrated standard physician's scales (Avery, 3302 ABN, England). Height was measured barefoot to the nearest centimetre using a portable stadiometer (Seca, Mod 220).

2.6 Maximum Voluntary Ventilation

Maximum voluntary ventilation ($\text{l}\cdot\text{min}^{-1}$) was performed using a clinical spirometer (Jaeger® Oxycon Pro®, Viasys health care, Version 5.2). The spirometer volume was calibrated prior each test using a standard 3l syringe. The testing procedure was explained and demonstrated to the volunteers immediately before the test and they were allowed a period of familiarisation. The procedure involves a short period of rapid and deep breathing (normally at a rate between 70-150 breaths per min and a depth between 0.25 and 0.75 of their vital capacity). It is important to emphasise that maximal effort is maintained for the entire measurement period (12s duration) so that two trials can be obtained with consistent effort and reproducibility. The higher value from the two trials is recorded.

2.7 Respiratory gas analysis

Continuous gas exchange measurements were made by open circuit spirometry (Jaeger® Oxycon Pro®, Viasys health care, Version 5.2). The accuracy and range of the relevant gas exchange variables from this system are illustrated in table

2.2 and the system was calibrated before each test in accordance with the manufacturer's instructions.

In summary, a sterilised turbine flow meter used for every test and the triple V turbine amplifier was calibrated by a manual pump with a defined volume (3l). This system has fast response differential chemo-O₂ and chemo-CO₂ analysers that are stable after a 15 minute warm-up. After this 15 minute delay, the appropriate analyser response was verified out by passing known gas mixture concentrations over the analysers. Inspired gas values were verified using ambient air (assumed to contain 20.90% O₂ and 0.03% CO₂) and appropriate expired gas values were verified using certified gas cylinders from the British Oxygen Corporation (containing 16.00±0.02% O₂ and 5.00±0.02% CO₂).

The system has no facility to externally adjust the amplifier gains on the O₂ and CO₂ analysers and such adjustment is made automatically. If the analyser gains cannot be adjusted to verify the known gas concentrations then they are replaced and the new analysers calibrated.

Variable	Range	Accuracy
Ventilation (l.min ⁻¹)	0 to 300	2% or 0.5
O ₂ uptake (l.min ⁻¹)	0 to 7	3% or 0.05
CO ₂ output (l.min ⁻¹)	0 to 7	3% or 0.05
RER	0.6 to 2.0	4%

Table 2.2 The range and accuracy of the common gas exchange variables as reported by the system manufacturer.

2.8 Familiarisation

Prior to testing the ramp protocols the volunteers were asked to attend familiarisation sessions within the laboratory environment. This allowed volunteers to meet the laboratory staff, observe the testing procedures and equipment and see the changing facilities. This is important psychological preparation for the subsequent testing sessions so that the volunteers are not distracted by their surroundings during data collection. Moreover, mini exercise testing sessions were conducted using both modalities so that the volunteer experienced the sensations associated with wearing and using the testing

equipment. In the current study at least one familiarisation can be enough to obtain reproducible and reliable (CPET) results (Mendez-Villanueva *et al.*, 2007).

During familiarisation, the communication (hand signals) and safety techniques (essentially how to stop a test in the event of an emergency) that are employed as standard operating procedures for (CPET) in our laboratories were explained. Obviously the insertion of a mouth piece and nose clip for respiratory gas analysis makes oral communication difficult and consequently communication with test participants is exclusively by hand signals. Volunteers were instructed that while they would be encouraged to make maximal efforts in all tests they could terminate the protocol at any time. Volunteers were encouraged to ask any questions that would help them feel comfortable during the test ramp protocol sessions.

2.9 Exercise modality and ramp protocols

The main aim of the current investigation aims to study the impact of changing exercise duration on maximal and sub-maximal markers of exercise capacity. As the current ACSM guidelines suggest that the exercise duration for (CPET) should lie between eight and twelve minutes it was hoped that ramp could be devised to allow volunteers to complete three ramp exercise durations. One with a short exercise duration (less than 8 minutes), one with a moderate exercise duration between (8 and 12 minutes) and one with long exercise duration (more than 12 minutes). It was further intended that the exercise durations could be applied in treadmill and cycle ergometer exercise; the two most common exercise modalities used in (CPET) throughout the world.

2.9.1 Treadmill ramp protocols

A considerable number of pilot treadmill studies were carried out to determine the protocols that would trigger maximal exertion with short, moderate and long exercise durations. These pilot studies were carried out by modifying the ramp Bruce protocol (Will & Walter, 1999). The ramp Bruce is a 60 work rate increment (CPET) protocol where the speed and/or gradient is changed every 15s. Using the original protocol and re-programming the duration of individual

work rate increments to 20s, 17s, 13s, 10s and 7s allowed us to examine which work rate increment duration might achieve the desired outcome of having three protocols to enable short, moderate and long exercise durations. Unfortunately this goal was not achievable in the heterogeneous population recruited and four treadmill protocols were required to bring the exercise durations close to the desired outcome (see figure 2.1).

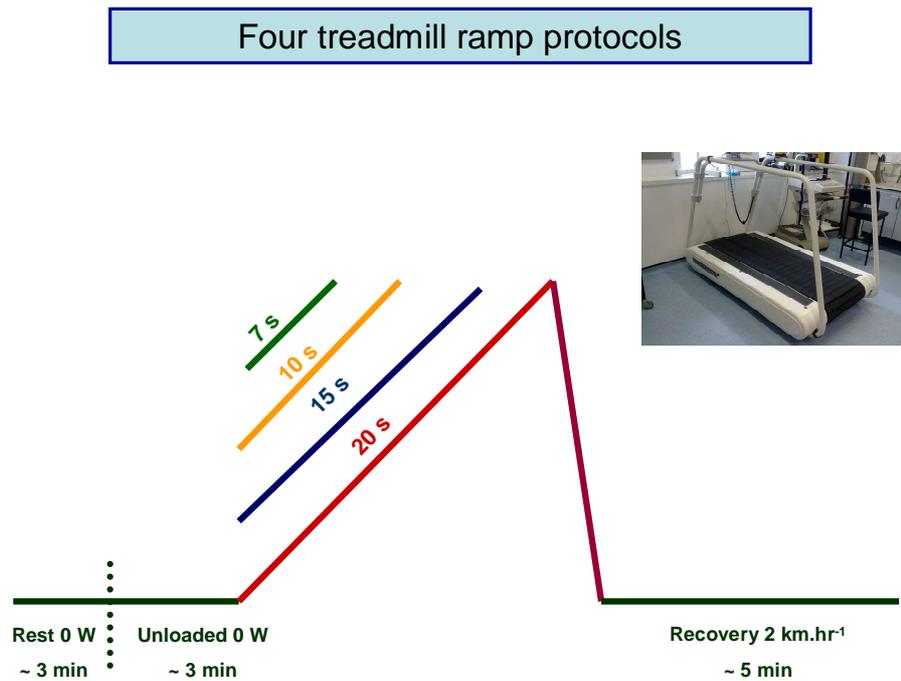


Figure 2-1 illustrates the experimental design used to assess the effects of exercise duration on markers of functional capacity during (CPET) on a treadmill. The green line represents 7 second work rate increments for a ramp Bruce protocol, the yellow 10 second work rate increments, the red 20 second work rate increments and the blue represents the standard ramp Bruce protocol.

As previously described the experimental design consisted of four stages; resting, unloaded, test ramp and recovery. The test ramp protocol was initiated after 3 minutes of unloaded walking. The speed and/or gradient was progressively increased according to the work rate increments within the ramp Bruce protocol (Will & Walter, 1999). However, the duration at which the work rate increments were maintained before progression to the next level was altered to influence the total exercise duration.

Changing the work rate increment every 7 seconds (green line in figure 2.1) or every 10 seconds (yellow line in figure 2.1) intervals resulted in a rapid increase in work rate so that volunteers reached their maximum effort in a relatively short time. These two work rate increment durations were required to allow short total exercise durations to be obtained in both high and low fitness volunteers. The standard ramp Bruce protocol (blue line in figure 2.1) was used to achieve moderate total exercise durations. Changing the work rate increment every 20 seconds (red line in figure 2.1) intervals resulted in a slow increase in work rate so that volunteers reached their maximum effort in a relatively longer time.

All participants were initially started with the 10 seconds work rate increment protocol to achieve short exercise durations. They were asked if they would return to complete a further 7 seconds work rate increment protocol if the initial test lasted longer than eight minutes. However, not all subjects were able to comply with this request within the required two weeks period for testing a specific exercise modality and consequently not all subjects were able to generate data that directly tests the ACSM guidelines.

However, even with this modification it is apparent that not all participants were able to match the desired total exercise duration using the standard or prolonged ramp Bruce protocol (see figure 2.2). This similar problem was encountered in the original study by Buchfuhrer and colleagues that forms the basis for the current ACSM recommendations on (CPET) (Buchfuhrer *et al.*, 1983). In this single study, 12 men completed various exercise tests (1- minute increment) to exhaustion on a treadmill and a cycle ergo meter. The breath by breath data from the best five subjects (mean age 36 ± 9.7 years) was analysed. It was concluded that the highest $\dot{V}_{O_2 \text{ MAX}}$ values were achieved for the five sub-groups between 8-17 minutes and with mean of 10 ± 2 minutes. In addition, the outcomes and recommendations were based just on a sub-group analysis of a small sample size, homogeneous group of fit young men (Buchfuhrer *et al.*, 1983). Even in this analysis, the prolongation of exercise duration did not significantly alter the maximum exercise response and the rationale for curtailing the exercise duration was merely on the convenience maximising appointments during a clinical exercise testing session. For this reason, no

further attempt was made to achieve the longer exercise durations desired and the main data analysis was performed in relation to 'within subject' variations in exercise duration before additional sub-group analysis of the best five volunteers fitting the ACSM guidelines.

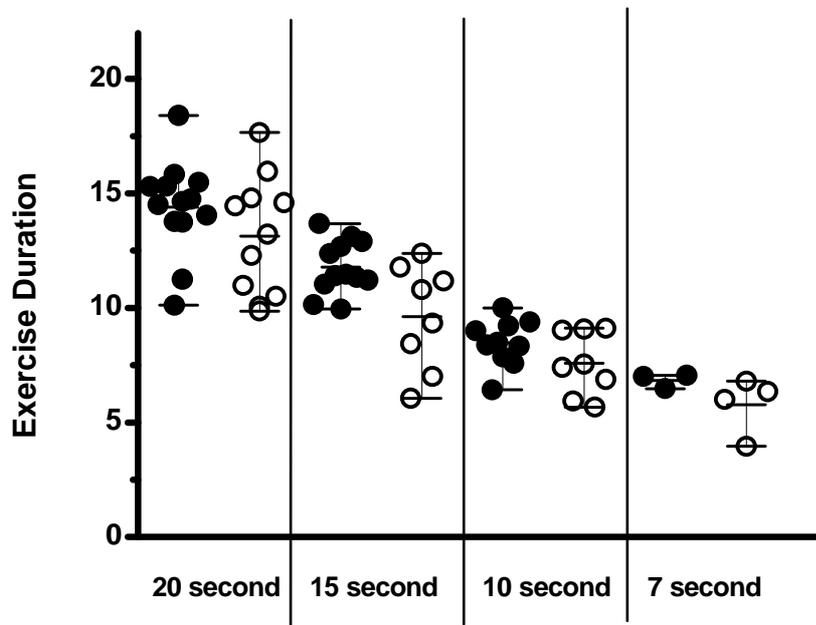


Figure 2-2 Pilot data showing the effects of changing the duration of the work rate increment on total exercise duration on a treadmill. Male volunteers are represented by closed circles and female volunteers by open circles

Treadmill testing was performed on a Woodway treadmill with a maximum speed of $20 \text{ km}\cdot\text{hr}^{-1}$ and maximum gradient of 20% (equivalent to $1:5$ or 11.31°). The accuracy of the indicated treadmill speed was measured as $\pm 0.1 \text{ km}\cdot\text{hr}^{-1}$ and maximum load-bearing capacity was 159 kg (Woodway GmbH Steinackerstr.20 operator Manual V0102, 1993).

The work load being performed during treadmill exercise can be calculated using the equation cited by Porszasz *et al* as follows (Porszasz *et al.*, 2003).

$$\text{WR} = m \cdot g \cdot v(t) \cdot \sin(\alpha) \quad (\text{eq 2.1})$$

where WR is the work in watts, m is body mass in kilogram, g is acceleration of a mass due to gravity = $9.81 \text{ m}\cdot\text{s}^{-2}$, v(t) is the time at the velocity, α is the angle of inclination.

2.9.2 Cycle ergometer ramp protocols

Pilot studies were also carried out on a cycle ergometer to determine protocols that would trigger maximal exertion with short, moderate and long exercise durations. These pilot studies were carried out using different work rate increment durations on a calibrated electronic cycle ergometer (Excalibur Sport, Lode BV, Gronigen, The Netherlands). A computer programmer was used to control the electromagnetic resistance on cycle ergometer wheel with range of workload from 10-1000W (equivalent to 60-6000 kpm.min⁻¹) with an accuracy $\leq 2\%$ from a starting work load resistance of 20W. During the familiarisation session, individual preparation for the cycle ergometer tests was completed. The height of seat and handlebars were adjusted to provide a comfortable riding position. These positions were recorded and were set for all the cycle ergometer test sessions.

As with the treadmill protocol design, the cycle ergometer tests consisted of four stages; resting, unloaded, test ramp and recovery. The test ramp protocol was initiated after 3 minutes of unloaded cycling. The rate of the increase in resistance applied to the flywheel was programmed and altered to influence the total exercise duration. Then unloaded stage was a constant 20W with a cadence of 45-55 revolutions per minute rpm. During the test ramp exercise, volunteers were asked to select a comfortable cadence between 60 and 100 rpm. All participants were instructed to remain seated on the cycle ergometer throughout the test. Active recovery was performed at a constant 20W and a 45-55 rpm cadence.

Four exercise ramp protocols on cycle ergometer

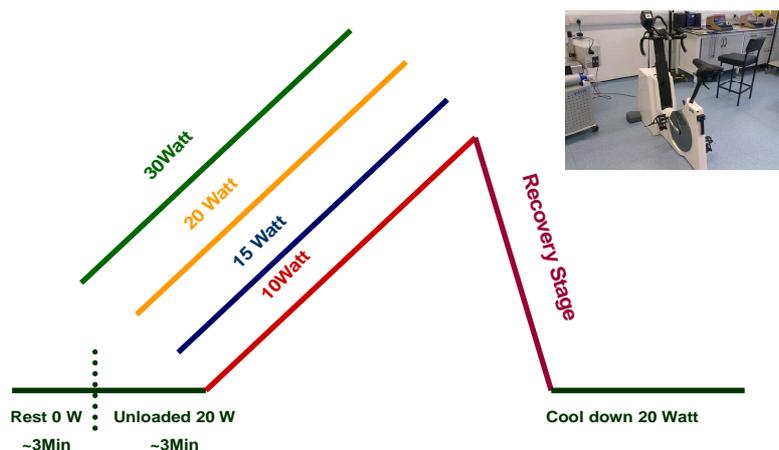


Figure 2-3 illustrates the experimental design used to assess the effects of exercise duration on markers of functional capacity during (CPET) on a cycle ergometer. The green line represents a $30 \text{ W}\cdot\text{min}^{-1}$ work rate increment for the ramp protocol, the yellow a $20 \text{ W}\cdot\text{min}^{-1}$ work rate increment, the blue a $15 \text{ W}\cdot\text{min}^{-1}$ work rate increment and the red a $10 \text{ W}\cdot\text{min}^{-1}$ work rate increment.

Increasing the resistance by $30 \text{ W}\cdot\text{min}^{-1}$ (green line in figure 2.3) or $20 \text{ W}\cdot\text{min}^{-1}$ (yellow line in figure 2.3) resulted in a rapid increase in work rate so that volunteers reached their maximum effort in a relatively short time. These two work rate increment durations were required to allow short total exercise durations to be obtained in both high and low fitness volunteers. The standard cycle ergometer ramp protocol employed in our laboratory of $15 \text{ W}\cdot\text{min}^{-1}$ resistance increments (blue line in figure 2.3) was used to achieve moderate total exercise durations. While a $10 \text{ W}\cdot\text{min}^{-1}$ work rate increment (red line in figure 2.3) resulted in a slow increase in work rate so that maximum effort was reached in a relatively longer time.

In a similar outcome to the treadmill studies, it was not possible to obtain three discrete protocols that met the aim of delivering short, moderate and long exercise durations that directly addressed the current ACSM guidelines on (CPET) (see figure 2.4). An additional protocol with a higher rate of resistance increase ($30 \text{ W}\cdot\text{min}^{-1}$) was included to try and achieve short exercise durations in high and low fitness volunteers and participants were asked to return if their initial $20 \text{ W}\cdot\text{min}^{-1}$ test did not meet that goal.

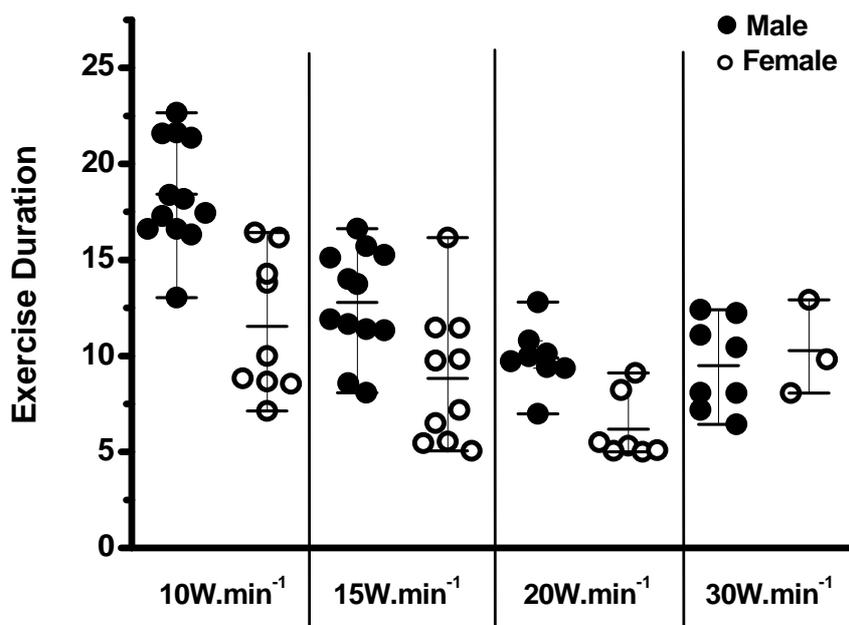


Figure 2-4 Pilot data showing the effects of changing the duration of the work rate increment on total exercise duration on a treadmill.

There was no further attempt to achieve the longer exercise durations desired and subsequently the main data analysis was performed in relation to 'within subject' variations in exercise duration before additional sub-group analysis of volunteers fitting the ACSM guidelines.

2.10 Heart Rate (HR)

Heart rate was collected throughout the experimental procedures every five seconds using a telemetry system (Garmin, Forerunner 50). The recorded heart rates were downloaded using Garmin software and analysed to determine peak heart rate recorded during the last 30 seconds of the test ramp protocol.

2.11 Maximal and sub-maximal markers of functional capacity

The aim of this programme of work is to extend the data available on the impact of exercise duration on markers of functional capacity during maximal (CPET). In addition, the effects of exercise duration will also be examined on sub-maximal indices that predict functional capacity and have diagnostic or prognostic value in clinical practice. The main indices will be discussed in detail

in introducing the relevant data chapters but a brief overview of the analytical procedures is given here.

2.12 Maximal indices of functional capacity

Breath by breath measurements of \dot{V}_{O_2} , \dot{V}_{CO_2} , fraction of end tidal O₂ concentration ($F_{ET}O_2\%$), fraction of end tidal CO₂ concentration ($F_{ET}CO_2\%$), \dot{V}_E and RER were made by open circuit spirometry using a previously described system. The noisy raw data was edited by applying a 95% confidence interval to a linear regression model and removing outlying data points using a commercially available data analysis and graphics package (Origin Pro Version 8, OriginLab Corporation, Northampton, USA). The indices of functional capacity at maximal effort are averaged from this edited data during the last 20 seconds of the test ramp protocol.

2.13 Ventilatory Anaerobic Threshold (VAT)

The edited data that is used to determine indices of functional capacity at maximum effort can then be used to establish the ventilatory anaerobic threshold VAT. The accuracy of the determination of the inflection point on the V-slope can be improved by editing the breath by breath raw data and averaging breaths (up to 10 breath averages) (Wasserman *et al.*, 1973; Beaver *et al.*, 1986). However, in most clinical testing situations the VAT is determined from raw data using the V-slope method and as such this will be the initial analysis applied here.

2.14 Oxygen Uptake Efficiency Slope (OUES)

As previously stated, the oxygen uptake efficiency slope (OUES) reflects the relationship between \dot{V}_{O_2} and \dot{V}_E during incremental exercise and is described by a single exponential function where the exponent gives the OUES. The OUES will be determined from the edited data used to determine maximal indices of functional capacity.

2.15 Breathing Reserve Index (BRI)

Breathing reserve index (BRI) is the fraction of maximal voluntary ventilation (MVV) used at peak exercise \dot{V}_E and is known as the peak breathing reserve index (BRI_{peak}). The determination of (MVV) has been described previously and peak \dot{V}_E will be analysed according to the methods for determining maximal indices of functional capacity. An alternative indices is the fraction of (MVV) used at the anaerobic threshold has been termed the breathing reserve index at anaerobic threshold (BRI_{AT}). The VAT for this analysis will be determined from raw data using the V-slope method.

2.16 Statistical analysis

The specific details of the statistical analysis applied to the data will be discussed as appropriate in each chapter. However, in general data are presented as mean \pm standard deviation and have been analysed using Origin Pro 8 SR4 v8. 0951(B951) and SPSS (18.0.0. Chicago). All data were normally distributed by Shapiro-Wilks normality test and suitable for parametric data analysis.

Multi-regression analyses are used for most independent variables that influence maximal and sub-maximal parameters with changing exercise duration. The agreement between maximal and sub-maximal indices during the comparison of moderate-short and moderate-long exercise durations are analysed by Bland and Altman tests.

CHAPTER THREE

Maximal Cardiopulmonary Measurements

3 Impact of Exercise Duration on Maximal Cardiopulmonary Measurements

3.1 Introduction

Cardiopulmonary exercise testing (CPET) is a common procedure used throughout the world in the diagnosis and prognosis of many medical conditions. It provides an objective method to evaluate the cardiopulmonary response of an individual to ramp or graded increment exercise most commonly using a treadmill or cycle ergometer (Franklin *et al.*, 2000; Mezzani *et al.*, 2009; ATS/ACCP, 2003; Stuart Jr & Ellestad, 1980). The most common forms of (CPET) stress the participant to their maximum effort using large muscle groups to try and obtain the maximum oxygen uptake ($\dot{V}_{O_2 \text{ MAX}}$) that can be measured.

The $\dot{V}_{O_2 \text{ MAX}}$ is the standard measure of aerobic capacity (the maximum rate at which an individual can take oxygen up from the air) and represents the integrated response of the pulmonary, cardiovascular and skeletal muscle systems to exercise. As previously discussed, some studies have suggested that the use of large work rate increments during (CPET) can cause the premature end to the test where muscle fatigue prevents a true measurement of $\dot{V}_{O_2 \text{ MAX}}$ (Buchfuhrer *et al.*, 1983). This means that the nature of the exercise protocol applied can influence the data generated at maximal effort and under these circumstances, where volitional exhaustion occurs before a plateau in \dot{V}_{O_2} , it can be more appropriate to use the term $\dot{V}_{O_2 \text{ PEAK}}$.

Changing the amplitude of work load increments during (CPET) can directly affect the exercise response. For example, during a standard Bruce protocol treadmill test the increase in speed and/or gradient every 3 minutes can be quite large and subsequently patients often cease exercise within a few seconds of the newly imposed work load. While the limiting factor in such cases is likely to be an inability to cope with the rapidly changing work load it is impossible to negate the concomitant influence on total exercise duration.

For this reason, the American College of Sports Medicine (ACSM) bases their guidelines for conducting clinical exercise testing around the duration of a test protocol. They state that an appropriate protocol should aim to have the participants reach $\dot{V}_{O_2\text{ PEAK}}$ within 8-12 minutes (Franklin *et al.*, 2000). However, the evidence supporting these guidelines is sparse and focused on \dot{V}_{O_2} as the outcome measure for aerobic capacity.

In reality, the diagnostic and prognostic power of (CPET) comes from interpretation of the integrated physiological response to exercise. Therefore, a range of exercise related parameters should be recorded to facilitate the process. So factors such as carbon dioxide output (\dot{V}_{CO_2}), minute ventilation (\dot{V}_E), the ventilatory equivalent for carbon dioxide ($\dot{V}_E / \dot{V}_{CO_2}$), respiratory exchange ratio RER and heart rate HR will be important like \dot{V}_{O_2} .

The aim of this study is, therefore, to add to the data that supports the current ACSM guidelines for clinical exercise testing. This additional data will not only increase the number of participants being studied, rather it will increase the relevance of the data for clinical populations by including women, older subjects and using the two most common exercise modalities. By using the same magnitude of work load increments and only changing the rate of change in the work load increment it will be possible to isolate the effects of exercise duration *per se*. Furthermore, the validity of the current guidelines will be tested in a sub-group analysis comparing maximal exercise responses in tests that comply with shorter and longer duration tests.

3.2 Methods

3.2.1 Subjects

The main aim of this study is to address the issue of exercise duration and therefore the analysis has been restricted to those volunteers that managed to complete at least three test ramp protocols on either the treadmill or cycle ergometer. Some of the volunteers managed to complete the necessary test ramp protocols on both modalities while others only managed this for one. The physical characteristics for the two groups are shown in table 3.1.

Variable	Gender	Treadmill	Cycle Ergometer
Number of Participants	Male	11	11
	Female	9	9
Age (years)	Male	31.2±6.0 (24-42)	32.5±6.0 (25-42)
	Female	36.6±11.0 * (24-57)	35.4±10.0 * (24-57)
Weight (kg)	Male	76.0±7.7 (60-84)	79.0±12.5 (60-103)
	Female	60.7±9.0 * (48-77)	60.0±9.0 * (48-77)
Height (cm)	Male	175.0±6.4 (163-182)	175.4±5.7 (163-182)
	Female	162.0±9.0 * (146-175)	160.7±7.3 * (145-172)
BSA [†] (m ²)	Male	1.92±0.14 (1.94-2.02)	1.96±0.12 (1.65-2.22)
	Female	1.65±0.14 * (1.47-1.87)	1.64±0.13 * (1.47-1.87)
BMI [#] (kg.m ²)	Male	24.8±2.0 (21-28)	26.0±4.0 (21-34)
	Female	23.1±3.8 * (18-29)	23.0±4.0 * (18-29)

Table 3.1 Physical characteristics of all study volunteers. Data are presented as mean±SD (where appropriate) and the range is given in parenthesis. * indicates $P<0.05$ compared with males (t -test with Bonferroni correction). [†] BSA is Body Surface Area according to Mosteller formula. [#] BMI is Body Mass Index.

3.3 Criteria for determination of peak oxygen uptake

Breath by breath measurement of \dot{V}_{O_2} was made by open circuit spirometry using a previously described system. The noisy raw data was edited by applying a 95% confidence interval to a linear regression model and removing outlying data points using a commercially available data analysis and graphics package (Origin Pro Version 8, Origin Lab Corporation, Northampton, USA). The \dot{V}_{O_2} at peak effort was averaged from this edited data during the last 20 seconds of the test ramp protocol. This procedure is illustrated in figure 3.1.

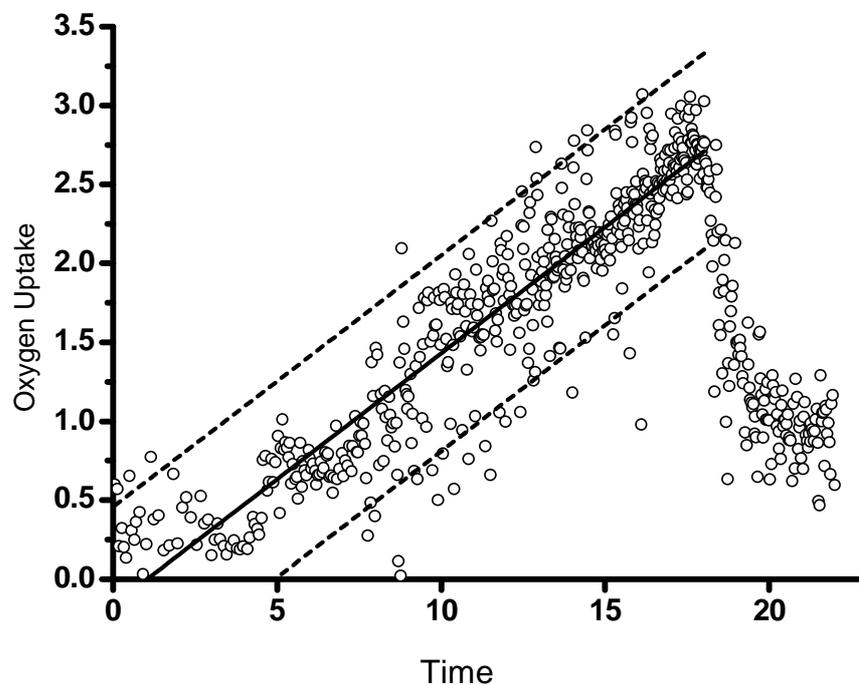


Figure 3-1 Illustrative example of how breath by breath raw data was edited. A linear regression model was applied to the raw data (solid line) and 95% confidence intervals applied (dashed lines). Any data lying outside the 95% confidence intervals was removed before averaging the oxygen uptake response from the edited data during the last twenty seconds of the test ramp protocol data.

3.4 Statistical Analysis

All data are presented as mean \pm standard deviation (SD) and were analyzed using Origin Pro 8 SR4 v8. 0951(B951), SPSS (18.0.0. Chicago) and MedCalc version 8.0.0.0. At the 0.05 level, all maximum variables are normally distributed by Shapiro-Wilks normality test and suitable for parametric data

analysis. All maximum variables were significantly drawn from a normal distributed population. A repeated measures was used to examine the differences in gas exchange parameters $\dot{V}_{O_2\text{ PEAK}}$, $\dot{V}_{E\text{ PEAK}}$, $\dot{V}_{CO_2\text{ PEAK}}$, $(\dot{V}_E / \dot{V}_{CO_2})$, $HR_{\text{ peak}}$ and maximal work load. If a significant was obtained, Bonferroni adjustments post hoc test was used for multiple comparisons between means. Statistical significance was set at 0.05 before Bonferroni adjustments.

The easy way to visualise limits of agreement and establish bias between two methods of measurement is Bland-Altman plots test. It is a bio-statistical method to calculate the mean difference between two methods of measurement (bias) agreement between two methods designed to measure the same parameter and were analysed by using MedCalc version 8.0.0.0 . And two-sample t-test utilised to compare the physical characteristics of the sample of the two exercise modality and if they likely from the same population.

A simple linear regression fits a straight line through the set of breath by breath points and it was edited by applying a 95% confidence interval to a linear regression model with upper and lower limit. The date out of upper and lower limit confidence interval was removed by using a commercially available data analysis and graphics package (Origin Pro Version 8, OriginLab Corporation, Northampton, USA).

3.5 Results

3.5.1 Physical characteristics

Table 3.1 indicates that the study participants are heterogeneous showing a wide range in age and body habitus. However, there was no significant difference in the physical characteristics between the two exercise modality groups. As expected, the male volunteers were significantly taller and heavier (resulting in higher BSA and BMI) than the female volunteers. The female group were significantly older than the males.

3.6 Influence of test ramp protocols on exercise duration

The different biomechanics and energy demands of walking and cycling can influence cardiopulmonary measurements during maximal effort. Consequently, the data for each exercise modality are presented separately.

3.6.1 Influence of test ramp protocols on treadmill exercise duration

Figure 3.2 shows the impact of short, moderate and long test ramp protocols (see figure 3.2 and associated text) on the total exercise duration achieved by the male and female volunteers. Males exercised for 8.0 ± 1.0 (range 6.4-9.4) minutes during the short protocol, 11.6 ± 1.1 (range 9.95-13.12) minutes during the moderate protocol and 14.4 ± 1.3 (range 11.25-15.83) minutes during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. The short protocol had significantly lower total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures) and the long protocol had significantly higher total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures). Females exercised for 6.8 ± 1.6 (range 4.0-9.1) minutes during the short protocol, 9.6 ± 2.0 (range 6.05-12.38) minutes during the moderate protocol and 13.0 ± 3.0 (range 9.86-17.66) minutes during the long protocol. Like their male counterparts, the short protocol had significantly lower total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures) and the long protocol had significantly higher total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures). There was no significant difference in

exercise duration between the male and female volunteers in the short, moderate or long test protocols.

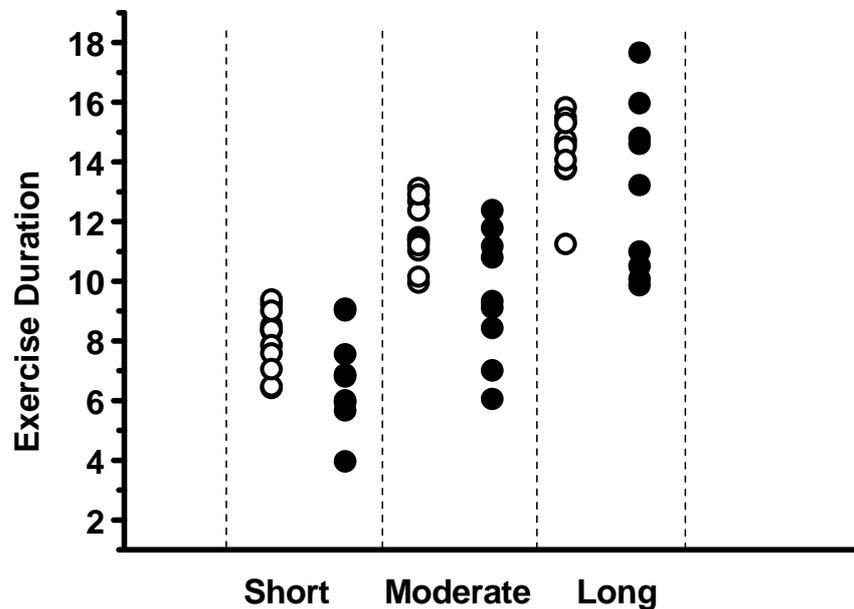


Figure 3-2 shows the effect of the test ramp treadmill protocols on exercise duration. Male volunteers are represented by open circles and female volunteers by closed circles. Exercise duration is in minutes and the short, moderate and long test ramp protocols are described in the methods section.

Influence of test ramp protocols on treadmill peak work load

Figure 3.3 shows the impact of short, moderate and long test ramp protocols (see figure 3.3 and associated text) on the peak work load achieved by the male and female volunteers. Males achieved peak work loads of 244 ± 51 (range 162-328) W during the short protocol, 215 ± 33 (range 167-270) W during the moderate protocol and 200 ± 28 (range 148.3-234) W during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the peak work rates achieved by the males in the test ramp protocols. Females achieved peak work loads of 169 ± 49 (range 118-249) W during the short protocol, 147 ± 47 (range 92-216) W during the moderate protocol and 142 ± 41 (range 102.5-211) minutes during the long protocol. Like their male counterparts, there was no significant difference in the peak work rates achieved by the females in the test ramp protocols. However, the female

peak work load was significantly lower than the male during the short and moderate test ramp protocols ($P < 0.05$, t -test).

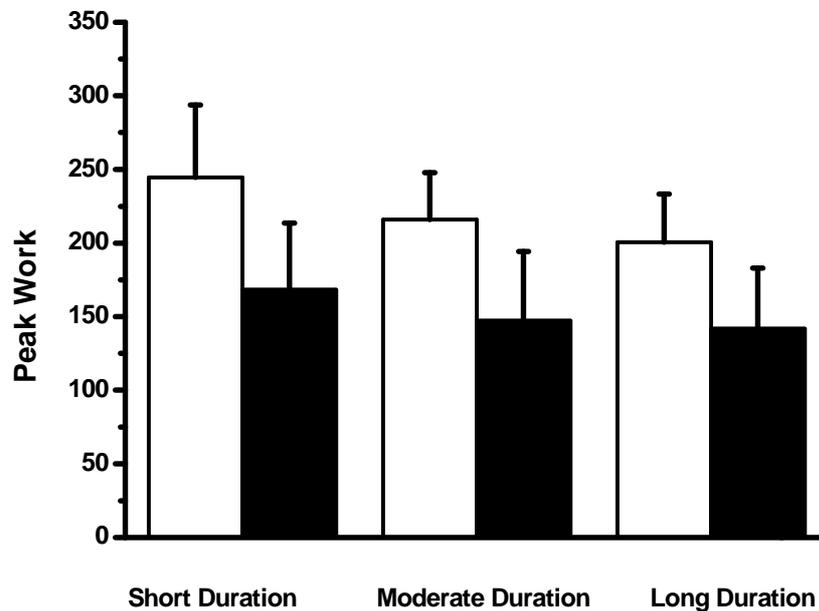


Figure 3-3 shows the effect of the test ramp treadmill protocols on peak work load. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak work is in watts and the short, moderate and long test ramp protocols are described in the methods section.

Influence of test ramp protocols on treadmill $\dot{V}_{O_2 \text{ PEAK}}$

Figure 3.4 shows the impact of short, moderate and long test ramp protocols (see figure 3.4 and associated text) on the $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the male and female volunteers. Males achieved an absolute $\dot{V}_{O_2 \text{ PEAK}}$ of 3.4 ± 0.6 (range 2.39-4.54) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 3.5 ± 0.5 (range 2.67-4.54) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 3.4 ± 0.5 (range 2.38-4.32) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the absolute $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the males in the test ramp protocols. Females achieved an absolute $\dot{V}_{O_2 \text{ PEAK}}$ of 2.4 ± 0.6 (range 1.65-3.3) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.3 ± 0.7 (range 1.48-3.38) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 2.4 ± 0.7 (range 1.55-3.63) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the females in the test ramp protocols.

However, the female absolute $\dot{V}_{O_2 \text{ PEAK}}$ was significantly lower than the male during the test ramp protocols ($P < 0.05$, t -test).

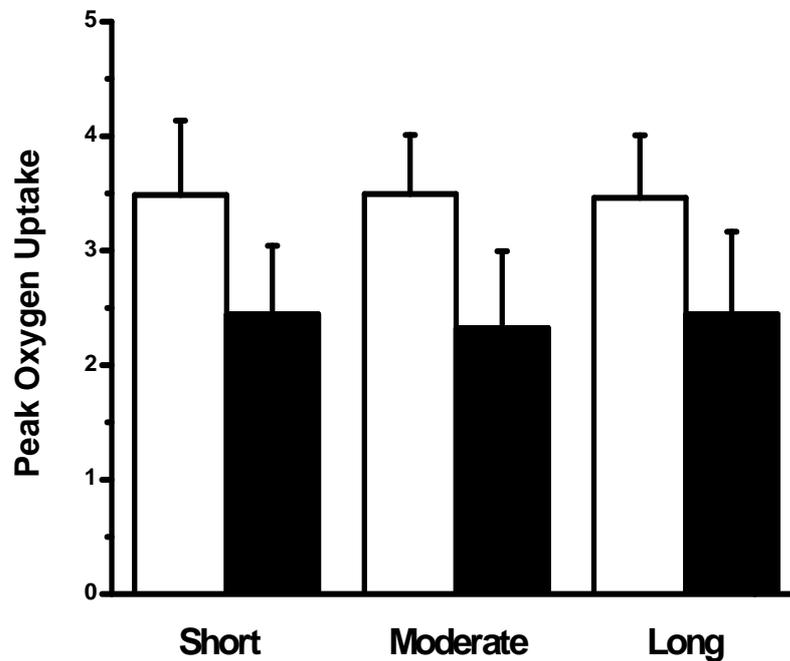


Figure 3-4 shows the effect of the test ramp treadmill protocols on $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Similarly, figure 3.5 shows that there was no difference in relative $\dot{V}_{O_2 \text{ PEAK}}$ when corrected for body weight. Males achieved a relative $\dot{V}_{O_2 \text{ PEAK}}$ of 45.0 ± 6.0 (range 31.4 -54) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the short protocol, 45.7 ± 5.5 (range 36.4-54) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the moderate protocol and 44.7 ± 5.0 (range 31.3-51.4) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the relative $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the males in the test ramp protocols. Females achieved a relative $\dot{V}_{O_2 \text{ PEAK}}$ of 38.9 ± 9.0 (range 29-52) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the short protocol, 38.0 ± 9.0 (range 25.1 -50.8) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the moderate protocol and 39.0 ± 9.0 (range 27.7-55.8) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the long protocol.

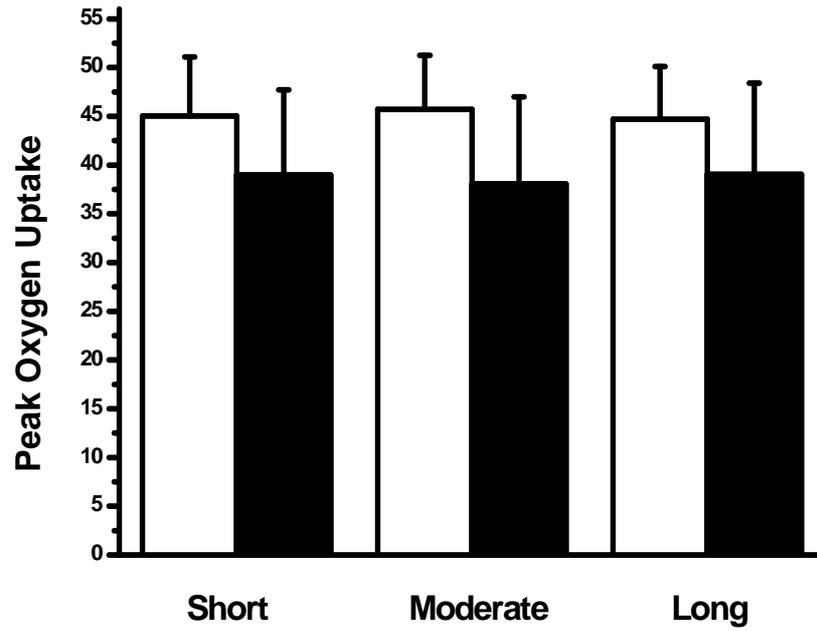


Figure 3-5 shows the effect of the test ramp treadmill protocols on $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in ml⁻¹.kg⁻¹.min⁻¹ and the short, moderate and long test ramp protocols are described in the methods section.

An example of the unedited raw data used to generate these results is shown in figure 3.6.

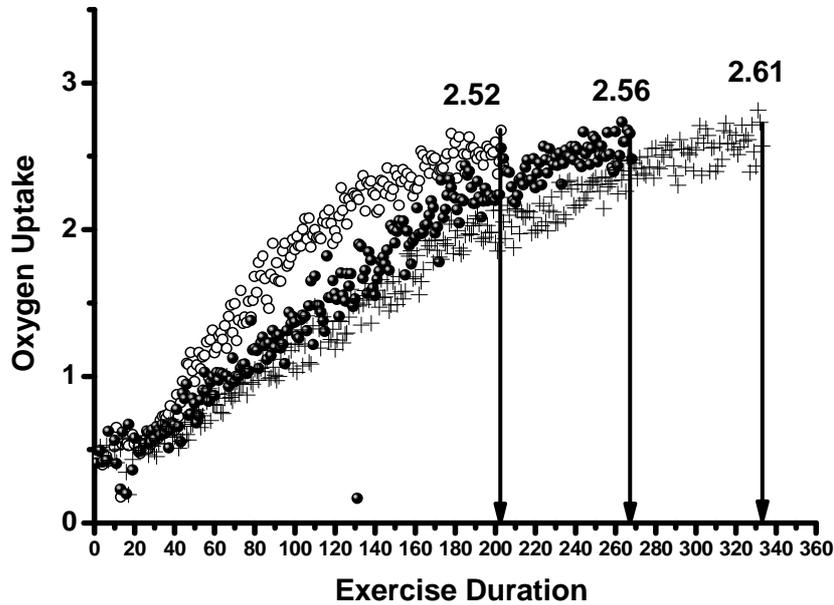


Figure 3-6 shows the effect of the test ramp treadmill protocols on raw \dot{V}_{O_2} data from an individual. Oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and exercise duration in seconds. In this case, all protocols elicit a $\dot{V}_{O_2 \text{ PEAK}}$ of around $2.5 \text{ l}\cdot\text{min}^{-1}$. The short (\circ), moderate (\bullet) and long ($+$) test ramp protocols are described in the methods section.

Bland-Altman plots are a bio-statistical method of analysing agreement between two methods designed to measure the same parameter. Although they are statistically inappropriate for testing agreement between different protocols applying the same methodology it is an easy way to visualise limits of agreement and establish bias. Figure 3.7 shows that despite relatively large standard deviations in the $\dot{V}_{O_2 \text{ PEAK}}$ measurements from the test ramp protocols there is good agreement across all the participants.

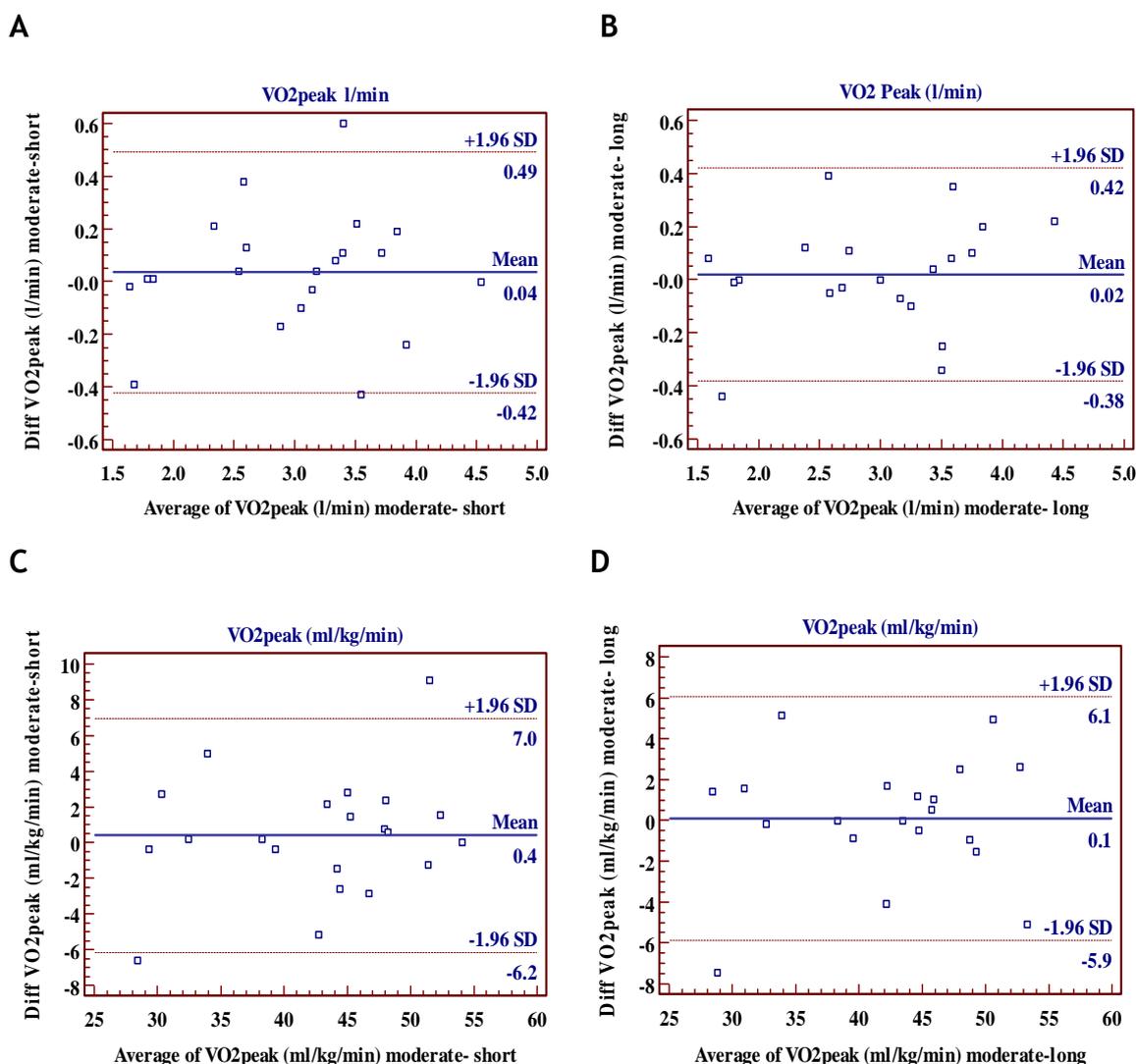


Figure 3-7 Bland-Altman plots showing agreement in $\dot{V}_{O_2 \text{ PEAK}}$ measurements across all study participants. The moderate test ramp protocol was used as the standard and plotted against the short or long test ramp protocols. Panel A illustrates absolute $\dot{V}_{O_2 \text{ PEAK}}$ with the short test ramp protocol and B the long test ramp protocol. Panel C illustrates relative $\dot{V}_{O_2 \text{ PEAK}}$ with the short test ramp protocol and D with the long test ramp protocol.

Influence of current ACSM guideline exercise duration on treadmill $\dot{V}_{O_2 \text{ PEAK}}$

The previous analysis reflected the impact of short, moderate and long test ramp treadmill protocols on exercise duration, peak work load and $\dot{V}_{O_2 \text{ PEAK}}$. However, not all participants attained the desired outcome where the short test ramp lasted less than eight minutes, the moderate between eight and twelve minutes and the long more than twelve minutes. While this previous analysis addresses the issue of exercise duration *per se*, it does not directly test the validity of the current ACSM guidelines. For this reason a further analysis has been made where individual tests are categorised for exercise duration rather than test ramp protocol (see table 3.2).

SN	sex	Time	$\dot{V}O_2$	Time	$\dot{V}O_2$	Time	$\dot{V}O_2$
1	F	7.55	1.83	10.8	1.84	13.22	1.84
2	F	9.08	2.52	12.38	2.56	15.96	2.61
3	F	5.66	1.87	7.00	1.48	10.07	1.92
4	F	9.02	2.97	11.78	2.80	14.80	2.69
5	F	3.95	2.23	6.05	2.44	9.86	2.32
6	F	6.87	1.78	9.33	1.79	10.98	1.80
7	F	5.93	1.65	8.43	1.63	10.50	1.55
8	F	6.80	3.30	9.11	3.38	17.66	3.63
9	F	6.00	3.10	11.17	3.00	14.60	3.00
10	M	9.22	3.66	12.68	3.77	13.75	3.42
11	M	8.50	3.40	11.47	3.62	14.65	3.54
12	M	8.33	3.16	11.40	3.20	15.83	3.30
13	M	8.38	2.54	11.33	2.67	14.75	2.70
14	M	7.85	3.16	11.03	3.13	13.78	3.20
15	M	9.38	4.04	12.37	3.80	15.32	3.70
16	M	6.42	2.39	9.95	2.77	11.25	2.38
17	M	9.00	3.75	13.12	3.94	15.48	3.74
18	M	6.47	3.34	11.21	3.45	14.51	3.41
19	M	7.58	3.76	10.15	3.33	14.05	3.67
20	M	7.05	4.54	12.90	4.54	15.30	4.32

Table 3.2 shows individual treadmill test $\dot{V}_{O_2 \text{ PEAK}}$ and exercise durations for all participants included in the initial analysis. The majority (43%) of tests conformed to the ACSM guidelines, with only 23% lasting less than eight minutes and 33% more than twelve minutes.

The exercise duration categorised data was then used to assess the validity of the ACSM guidelines by plotting the differences between absolute $\dot{V}_{O_2 \text{ PEAK}}$ measured in tests that conformed to those guidelines and those shorter or longer.

Figure 3.8 shows that there was no significant difference in absolute $\dot{V}_{O_2 \text{ PEAK}}$.

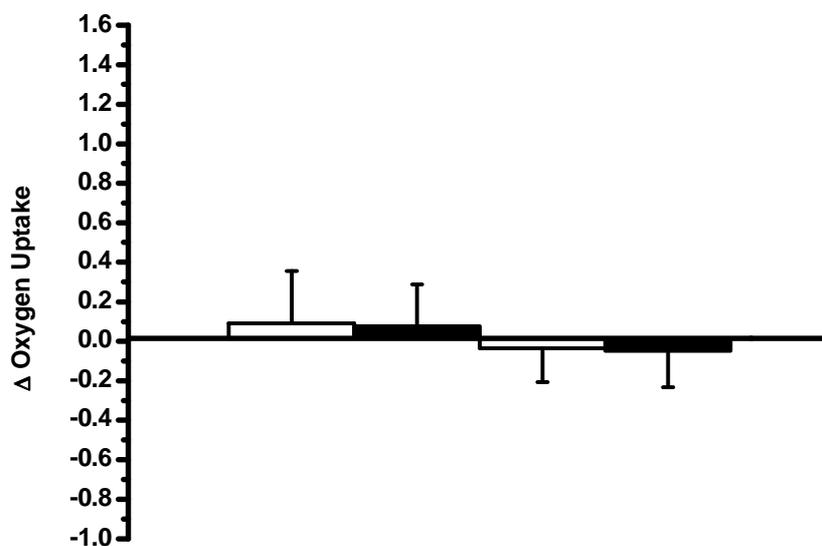


Figure 3-8 shows the effect of exercise duration not conforming to the current ACSM guidelines on absolute $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and the first two columns show the difference in absolute oxygen consumption when exercise was shorter than the ACSM guidelines. The last two columns show the difference in absolute oxygen consumption when exercise was longer than the ACSM guidelines.

Similarly, figure 3.9 shows that there was no significant difference in relative

$$\dot{V}_{O_2\text{PEAK}}$$

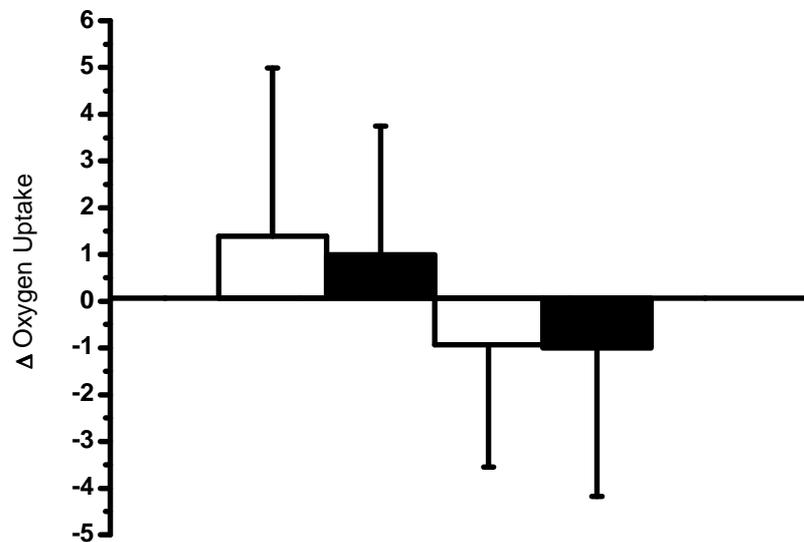


Figure 3-9 shows the effect of exercise duration not conforming to the current ACSM guidelines on relative $\dot{V}_{O_2\text{PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in $\text{ml}^{-1} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ and the first two columns show the difference in absolute oxygen consumption when exercise was shorter than the ACSM guidelines. The last two columns show the difference in absolute oxygen consumption when exercise was longer than the ACSM guidelines.

Influence of test ramp protocols on other treadmill peak parameters

Figure 3.10 shows the impact of short, moderate and long test ramp protocols (see figure 3.10 and associated text) on the $\dot{V}_{\text{CO}_2\text{PEAK}}$ achieved by the male and female volunteers. Males achieved $\dot{V}_{\text{CO}_2\text{PEAK}}$ of 4.0 ± 0.7 (range 2.82-5.42) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 4.0 ± 0.6 (range 3.36-4.95) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 3.9 ± 0.5 (range 2.72-4.42) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the $\dot{V}_{\text{CO}_2\text{PEAK}}$ achieved by the males in the test ramp protocols. Females achieved $\dot{V}_{\text{CO}_2\text{PEAK}}$ of 2.8 ± 0.7 (range 1.83-3.66) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.6 ± 0.7 (range 1.5-3.4) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol

and 2.6 ± 0.7 (range 1.6-3.82) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ achieved by the females in the test ramp protocols.

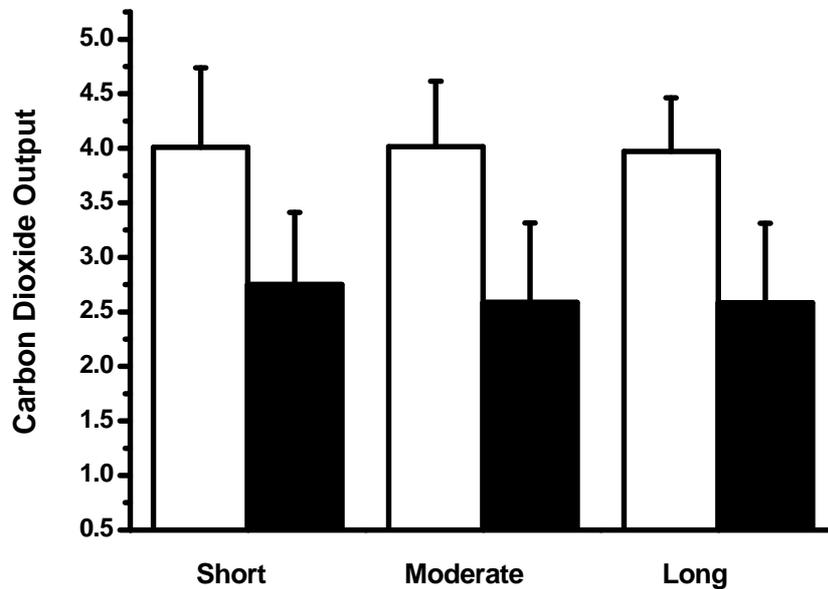


Figure 3-10 shows the effect of the test ramp treadmill protocols on $\dot{V}_{\text{CO}_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak carbon dioxide output is in $\text{l} \cdot \text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Figure 3.11 shows the impact of short, moderate and long test ramp protocols (see figure 3.11 and associated text) on the $\dot{V}_{\text{E PEAK}}$ achieved by the male and female volunteers. Males achieved $\dot{V}_{\text{E PEAK}}$ of 118 ± 24 (range 82.3-161) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 118 ± 19 (range 95-153.4) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 114 ± 22 (range 78.1-146) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the $\dot{V}_{\text{E PEAK}}$ achieved by the males in the test ramp protocols. Females achieved $\dot{V}_{\text{E PEAK}}$ of 88 ± 19 (range 55.8-115) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 80 ± 18 (range 53-94) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 83 ± 21 (range 50-112) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male

counterparts, there was no significant difference in $\dot{V}_{E\text{ PEAK}}$ achieved by the females in the test ramp protocols.

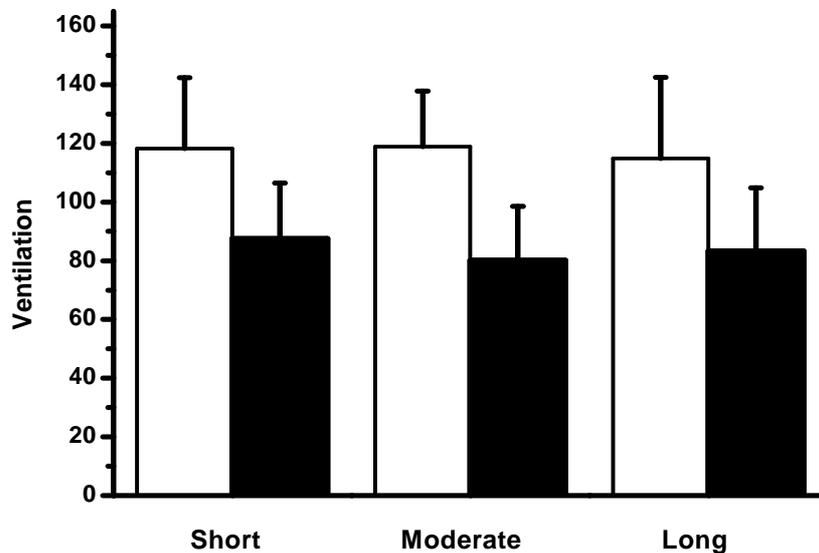


Figure 3-11 shows the effect of the test ramp treadmill protocols on $\dot{V}_{E\text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak ventilation is in $\text{l}\cdot\text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Figure 3.12 shows the impact of short, moderate and long test ramp protocols (see figure 3.12 and associated text) on the $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the male and female volunteers. Males achieved a $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio of 28.8 ± 4 (range 23.8-35.5) during the short protocol, 30 ± 3 (range 24.2-34.3) during the moderate protocol and 30 ± 4 (range 22.4-37.2) during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the males in the test ramp protocols. Females achieved a $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio of 32 ± 3 (range 27.2-36.2) during the short protocol, 31 ± 4 (range 27.5-36.6) during the moderate protocol and 32 ± 4 (range 27-40) during the long protocol. Like their male counterparts, there was no significant difference in $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the females in the test ramp protocols.

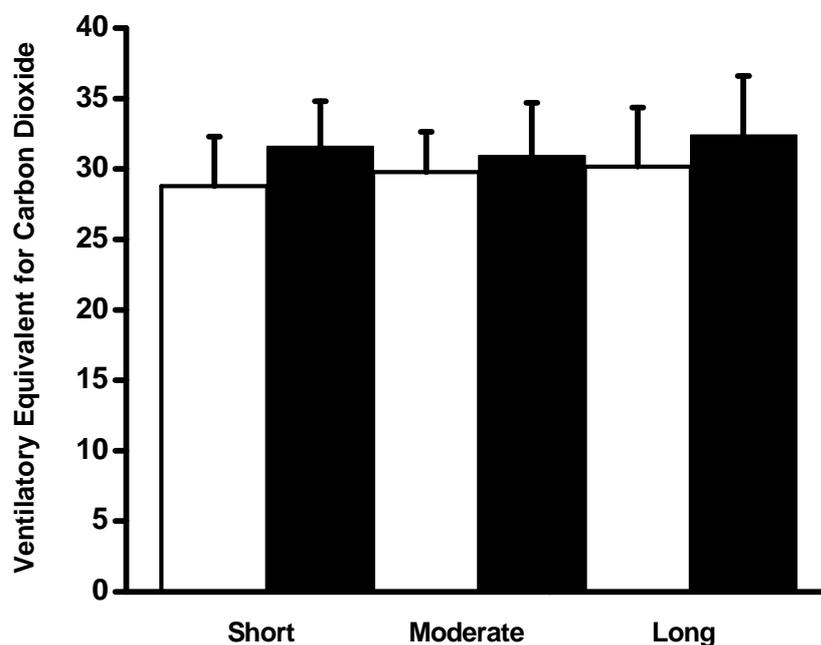


Figure 3-12 shows the effect of the test ramp treadmill protocols on $\dot{V}_E / \dot{V}_{CO_2}$ ratio. Male volunteers are represented by the open bars and female volunteers by the closed bars. The short, moderate and long test ramp protocols are described in the methods section.

As stated previously, Bland-Altman plots are statistically inappropriate for testing agreement between different protocols applying the same methodology but have been applied to visualise limits of agreement and establish bias. Figure 3.12 shows that there is good agreement in various parameters across all the participants.

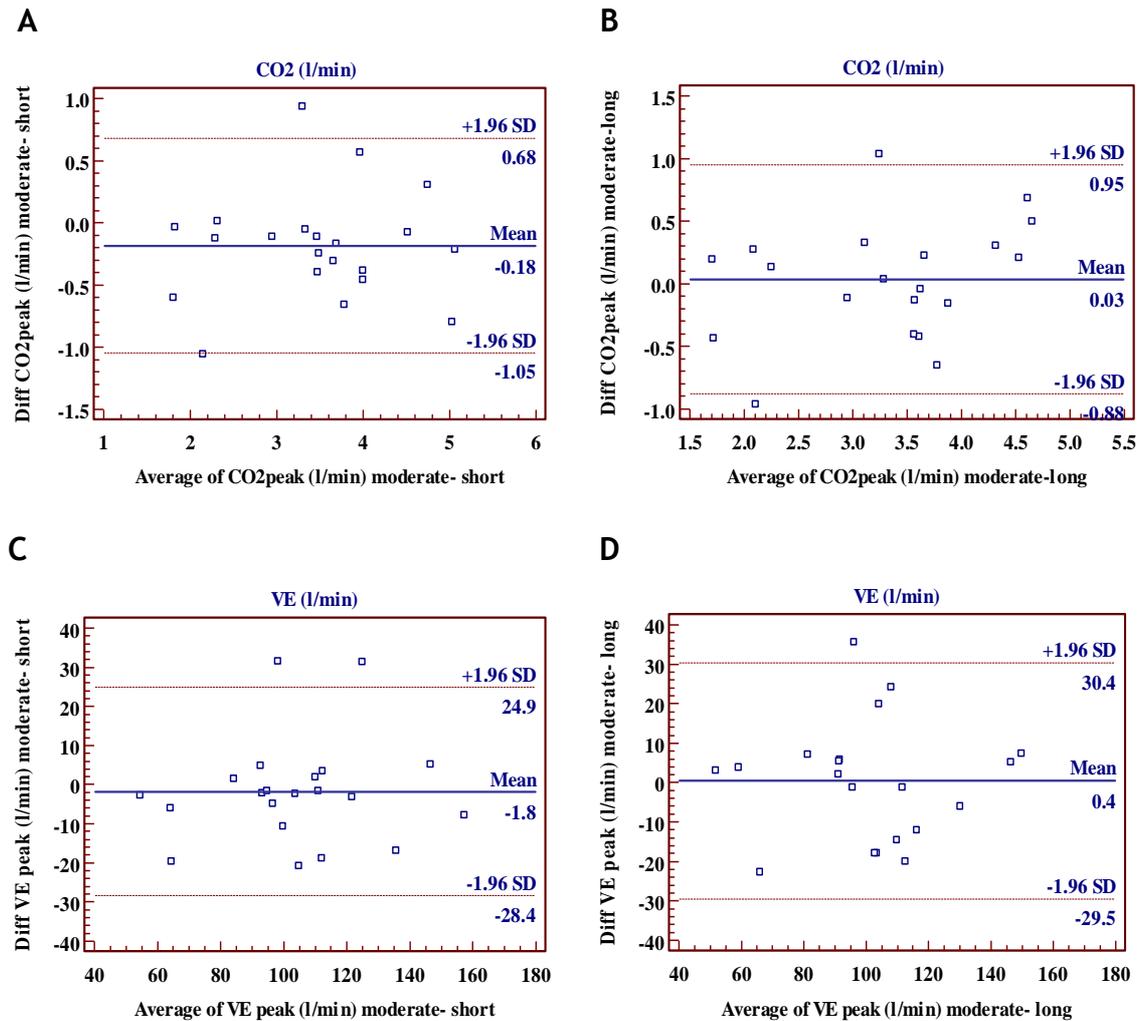


Figure 3-13 Bland-Altman plots showing agreement in $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ and \dot{V}_E measurements across all study participants. The moderate test ramp protocol was used as the standard and plotted against the short or long test ramp protocols. Panel A illustrates absolute $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ with the short test ramp protocol and B the long test ramp protocol. Panel C illustrates \dot{V}_E with the short test ramp protocol and D with the long test ramp protocol.

Figure 3.14 shows the impact of short, moderate and long test ramp protocols (see figure 3.14 and associated text) on the peak RER achieved by the male and female volunteers. Males achieved a peak RER of 1.23 ± 0.11 (range 1.08-1.48) during the short protocol, 1.17 ± 0.11 (range 1.03-1.35) during the moderate protocol and 1.14 ± 0.10 (range 0.99-1.34) during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. The peak RER achieved by the males in the short test ramp protocol was significantly higher than in the long test ramp protocol ($P < 0.01$ by repeated measures). Females achieved a peak RER of 1.17 ± 0.10 (range 1.04-1.31) during the short protocol, 1.13 ± 0.1 (range 1-1.26) during the moderate protocol and 1.10 ± 0.10 (range 1-

1.19) during the long protocol. There was no significant difference in peak RER achieved by the females in the test ramp protocols.

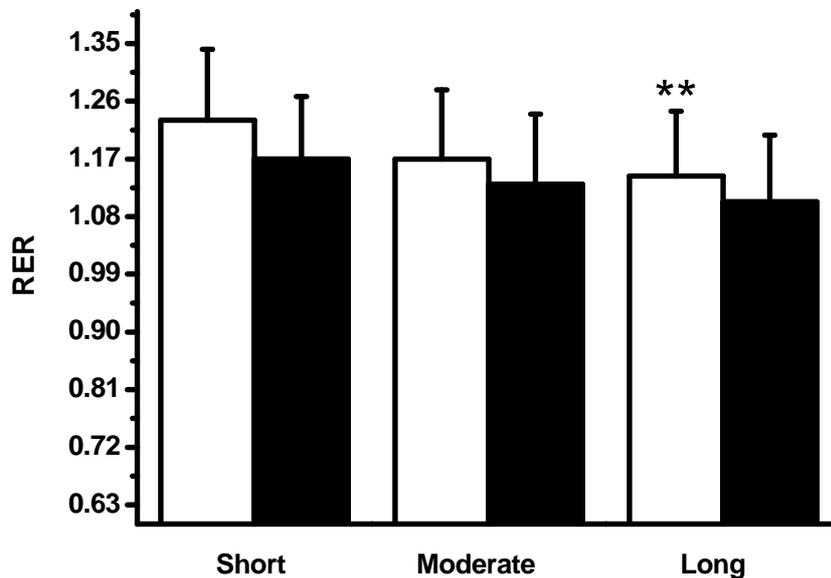


Figure 3-14 shows the effect of the test ramp treadmill protocols on peak RER. Male volunteers are represented by the open bars and female volunteers by the closed bars. The short, moderate and long test ramp protocols are described in the methods section. ** $P < 0.01$ compared with the short test ramp protocol in males.

Due to technical problems with the heart rate monitors during some of the test ramp protocols, heart rate data from three female and one male volunteers were excluded from data analysis. Figure 3.15 shows the impact of short, moderate and long test ramp protocols (see figure 3.15 and associated text) on the peak heart rate achieved by the male and female volunteers. Males achieved a peak heart rate of 187 ± 8 (range 173-202) during the short protocol, 183 ± 11 (range 166-203) during the moderate protocol and 185 ± 9 (range 169-200) during the long protocol. One male subject's data is excluded from the short protocol because he managed to complete all 60 work load increments without exhaustion. There was no significant difference in the peak heart rate achieved by the males in the test ramp protocols. Females achieved a peak heart rate of 179 ± 12 (range 158-192) during the short protocol, 180 ± 11 (range 167-201) during the moderate protocol and 179 ± 10 (range 165-194) during the long protocol. Like their male counterparts, there was no significant difference in peak heart rate achieved by the females in the test ramp protocols.

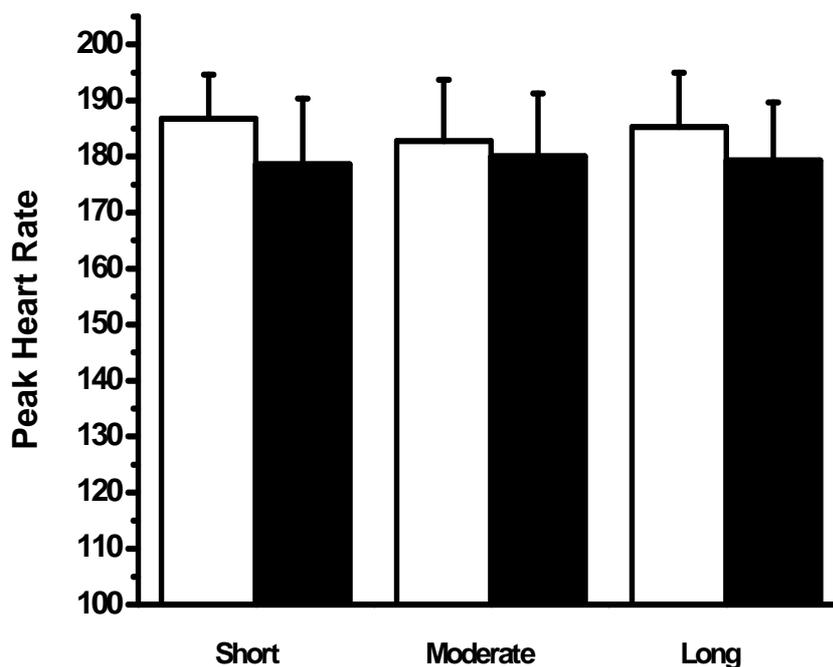


Figure 3-15 shows the effect of the test ramp treadmill protocols on peak heart rate. Male volunteers are represented by the open bars and female volunteers by the closed bars. Heart rate is in beats per minute and the short, moderate and long test ramp protocols are described in the methods section.

3.6.2 Influence of test ramp protocols on cycle ergometer exercise duration

Figure 3.16 shows the impact of short, moderate and long test ramp protocols (see figure 3.16 and associated text) on the total exercise duration achieved by the male and female volunteers. Males exercised for 10.00 ± 2.00 (range 6.43-12.80) minutes during the short protocol, 12.30 ± 3.00 (range 6.98-15.7) minutes during the moderate protocol and 17.11 ± 4.00 (range 8.08-21.62) minutes during the long protocol. The short protocol had significantly lower total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures) and the long protocol had significantly higher total exercise duration than the moderate protocol ($P < 0.05$, by repeated measures). Females exercised for 6.80 ± 2.00 (range 5-9.81) minutes during the short protocol, 8.00 ± 2.60 (range 5.06-11.48) minutes during the moderate protocol and 11.54 ± 4.00 (range 7.13-16.43) minutes during the long protocol.

minutes during the long protocol. Like their male counterparts, the short protocol had significantly lower total exercise duration than the moderate protocol ($P<0.05$, by repeated measures) and the long protocol had significantly higher total exercise duration than the moderate protocol ($P<0.05$, by repeated measures). The female exercise durations were significantly lower than the males for all test protocols ($P<0.01$ in all cases, by repeated measures).

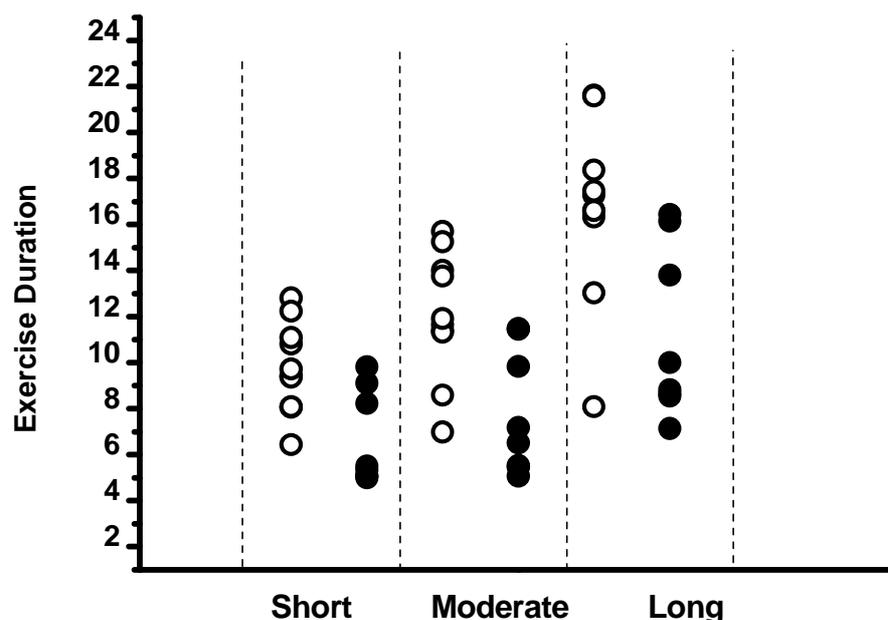


Figure 3-16 shows the effect of the test ramp treadmill protocols on exercise duration. Male volunteers are represented by open circles and female volunteers by closed squares. Exercise duration is in minutes and the short, moderate and long test ramp protocols are described in the methods section.

Influence of test ramp protocols on cycle ergometer peak work load

Figure 3.17 shows the impact of short, moderate and long test ramp protocols (see figure 3.17 and associated text) on the peak work load achieved by the male and female volunteers. Males achieved peak work loads of 277 ± 68 (range 207-393) W during the short protocol, 208.2 ± 36 (range 149-256) W during the moderate protocol and 193.1 ± 29 (range 142-236) W during the long protocol. Their peak work rates achieved by the males was significantly higher in the short test ramp protocol ($P<0.01$, by repeated measures). Females achieved peak work loads of 173 ± 69 (range 120-315) W during the short protocol, 140.8 ± 38

(range 96-192) W during the moderate protocol and 135.9 ± 35 (range 91-184) minutes during the long protocol. There was no significant difference in the peak work rates achieved by the females in the test ramp protocols. Female peak work load was significantly lower than the male during all test ramp protocols ($P < 0.05$, by repeated measures).

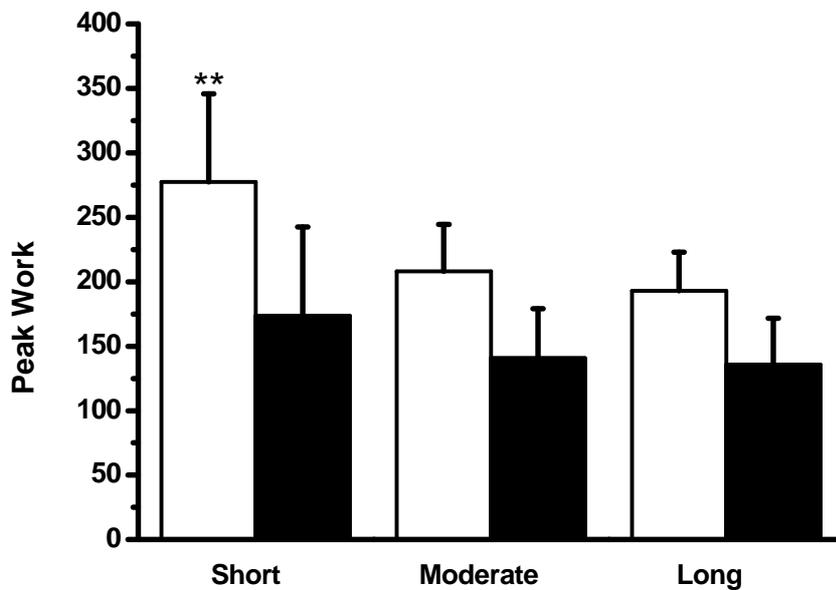


Figure 3-17 shows the effect of the test ramp cycle ergometer protocols on peak work load. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak work is in watts and the short, moderate and long test ramp protocols are described in the methods section. ** $P < 0.01$ compared with the moderate and long test ramp protocols.

Influence of test ramp protocols on cycle ergometer $\dot{V}_{O_2 \text{ PEAK}}$

Figure 3.18 shows the impact of short, moderate and long test ramp protocols (see figure 3.18 and associated text) on the $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the male and female volunteers. Males achieved an absolute $\dot{V}_{O_2 \text{ PEAK}}$ of 3.0 ± 0.5 (range 2.0-3.7) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 3.0 ± 0.5 (range 2.2-3.5) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 3.0 ± 0.6 (range 2.0-3.8) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the males in the test ramp protocols. Females achieved an absolute $\dot{V}_{O_2 \text{ PEAK}}$ of 1.9 ± 0.5 (range 1.4-2.8) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.9 ± 0.4 (range 1.34-2.57) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 2.0 ± 0.5 (range 1.31-2.67) $\text{l} \cdot \text{min}^{-1}$ during the

long protocol. Like their male counterparts, there was no significant difference in the absolute $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the females in the test ramp protocols. However, the female absolute $\dot{V}_{O_2 \text{ PEAK}}$ was significantly lower than the male during the test ramp protocols ($P < 0.01$, by repeated measures).

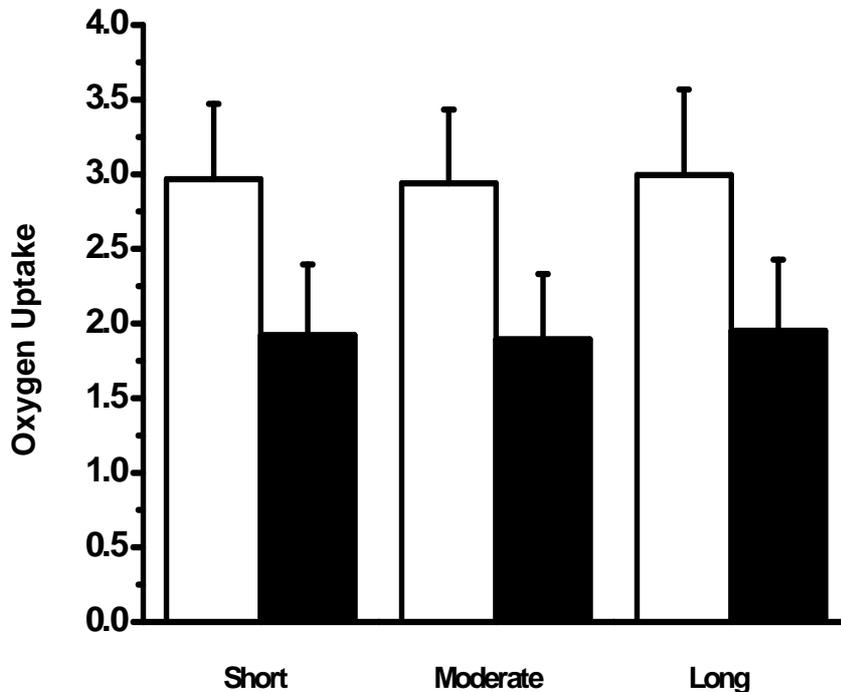


Figure 3-18 shows the effect of the test ramp cycle ergometer protocols on $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Similarly, figure 3.19 shows that there was no difference in relative $\dot{V}_{O_2 \text{ PEAK}}$ when corrected for body weight. Males achieved a relative $\dot{V}_{O_2 \text{ PEAK}}$ of 38.7 ± 7.4 (range 29.1-49.2) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the short protocol, 37.7 ± 6.7 (range 27.8-50.6) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the moderate protocol and 38.1 ± 5.9 (range 28.4-45.4) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the long protocol. There was no significant difference in the relative $\dot{V}_{O_2 \text{ PEAK}}$ achieved by the males in the test ramp protocols. Females achieved a relative $\dot{V}_{O_2 \text{ PEAK}}$ of 31.6 ± 6.3 (range 24.7-40.4) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the short protocol, 31.4 ± 7.2 (range 22.9-43.8) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the moderate protocol and 32.1 ± 6.9 (range 22.2-42.3) $\text{ml}^{-1}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ during the long protocol.

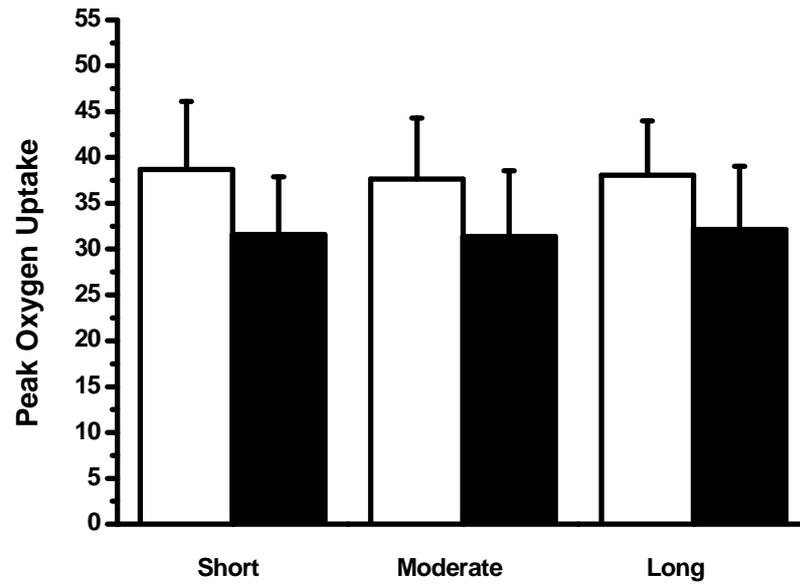


Figure 3-19 shows the effect of the test ramp cycle ergometer protocols on $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in ml⁻¹.kg⁻¹.min⁻¹ and the short, moderate and long test ramp protocols are described in the methods section.

An example of the unedited raw data used to generate these results is shown in figure 3.20.

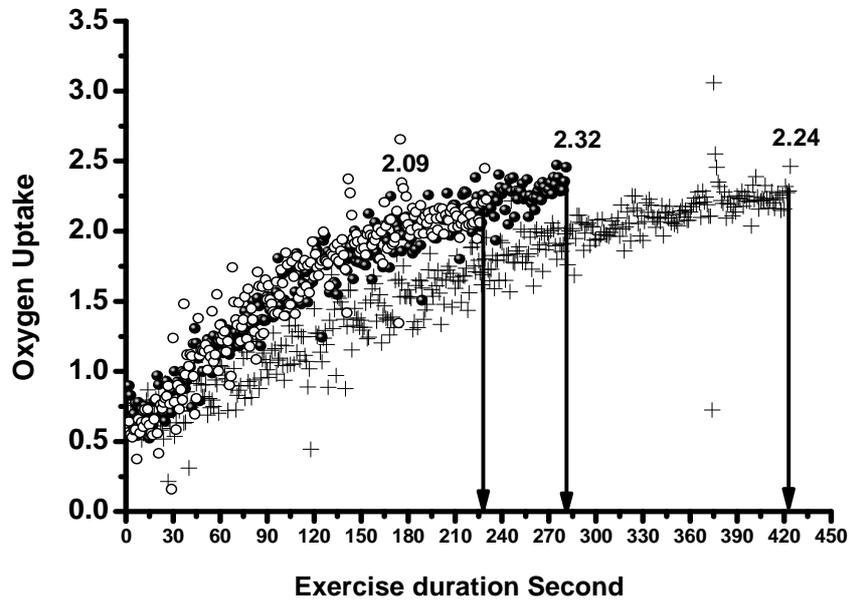


Figure 3-20 shows the effect of the test ramp cycle ergometer protocols on raw $\dot{V}_{O_{2, \text{PEAK}}}$ data from an individual. Oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and exercise duration in seconds. In this case, all protocols elicit a $\dot{V}_{O_{2, \text{PEAK}}}$ of around $2.2 \text{ l}\cdot\text{min}^{-1}$. The short (\circ), moderate ($*$) and long ($+$) test ramp protocols are described in the methods section.

As stated previously, Bland-Altman plots are statistically inappropriate for testing agreement between different protocols applying the same methodology but have been applied to visualise limits of agreement and establish bias. Figure 3.21 shows that there is good agreement in various parameters across all the participants.

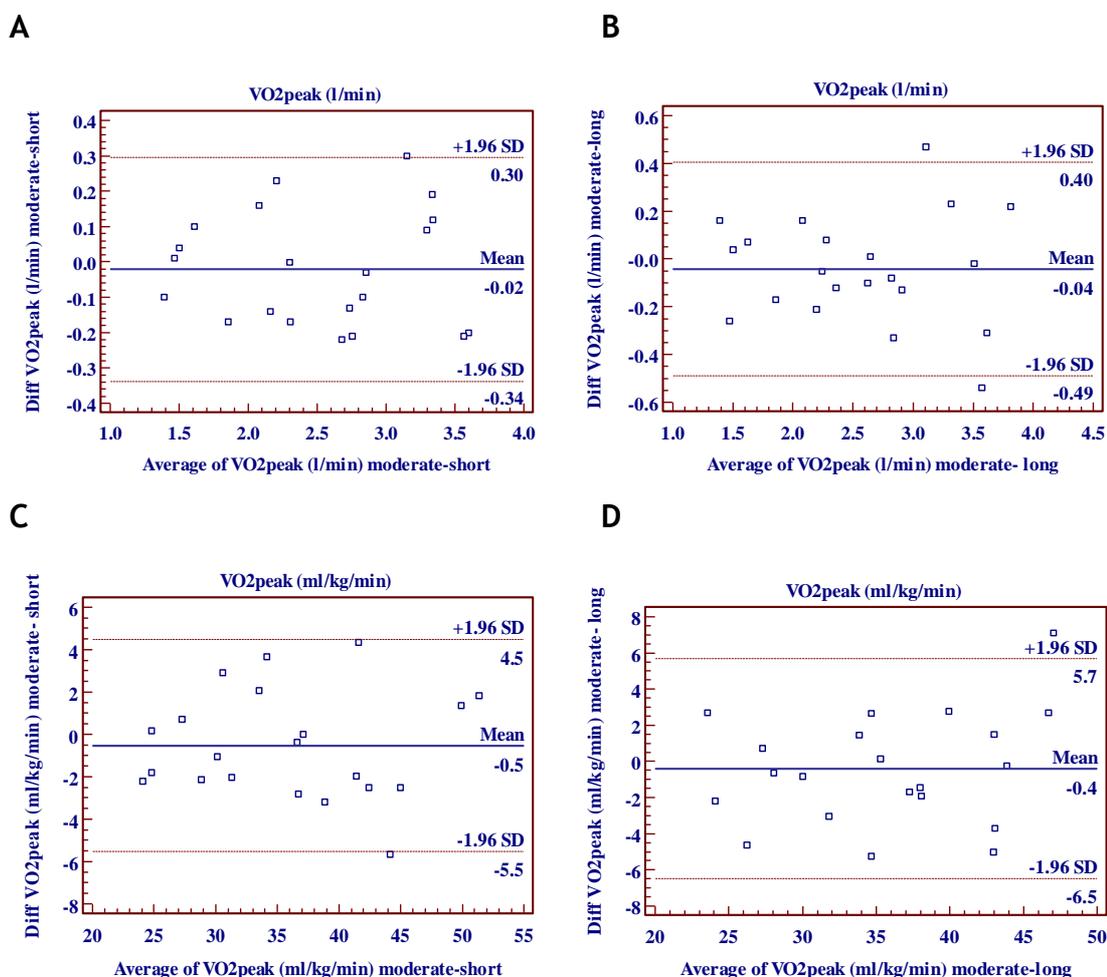


Figure 3-21 Bland-Altman plots showing agreement in $\dot{V}_{O_2 \text{ PEAK}}$ measurements across all study participants. The moderate test ramp protocol was used as the standard and plotted against the short or long test ramp protocols. Panel A illustrates absolute $\dot{V}_{O_2 \text{ PEAK}}$ with the short test ramp protocol and B the long test ramp protocol. Panel C illustrates relative $\dot{V}_{O_2 \text{ PEAK}}$ with the short test ramp protocol and D with the long test ramp protocol.

Influence of current ACSM guideline exercise duration on cycle ergo meter

$$\dot{V}_{O_2 \text{ PEAK}}$$

The previous analysis reflected the impact of short, moderate and long test ramp cycle ergometer protocols on exercise duration, peak work load and $\dot{V}_{O_2 \text{ PEAK}}$. However, not all participants attained the desired outcome where the short test ramp lasted less than eight minutes, the moderate between eight and twelve minutes and the long more than twelve minutes. While this previous analysis addresses the issue of exercise duration *per se*, it does not directly test the validity of the current ACSM guidelines. For this reason a further analysis has been made where individual tests are categorised for exercise duration rather than test ramp protocol (see table 3.3).

SN	sex	Time	$\dot{V}O_2$	Time	$\dot{V}O_2$	Time	$\dot{V}O_2$
1	F	5.33	1.56	7.17	1.66	10.00	1.59
2	F	9.10	2.09	11.45	2.32	16.15	2.24
3	F	5.06	1.46	5.53	1.47	7.13	1.31
4	F	8.22	2.23	9.83	2.09	13.8	2.30
5	F	5.00	1.94	5.45	1.77	8.66	1.94
6	F	5.50	1.48	6.50	1.52	8.83	1.48
7	F	5.08	1.44	5.06	1.34	8.55	1.60
8	F	9.81	2.79	11.48	2.57	16.43	2.67
9	F	8.06	2.30	9.75	2.30	14.28	2.42
10	M	9.42	2.87	11.40	2.84	16.60	2.97
11	M	8.06	2.80	11.65	2.67	17.28	3.00
12	M	9.37	2.88	11.33	2.78	16.32	2.86
13	M	6.43	2.00	6.98	2.16	8.08	2.00
14	M	8.08	2.39	8.58	2.22	13.03	2.27
15	M	12.8	3.70	15.70	3.50	21.62	3.52
16	M	9.72	2.86	11.91	2.65	16.6	2.64
17	M	10.80	3.24	14.00	3.43	18.37	3.20
18	M	11.08	3.25	13.75	3.34	17.45	2.87
19	M	12.23	3.67	15.25	3.46	21.58	3.77
20	M	12.41	3.00	15.11	3.30	21.35	3.84

Table 3.3 shows individual cycle ergometer test $\dot{V}O_{2\text{ PEAK}}$ and exercise durations for all participants included in the initial analysis. Many of the tests (41%) conformed to the ACSM guidelines, with only 22% lasting less than eight minutes and 37% more than twelve minutes.

The exercise duration categorised data was then used to assess the validity of the ACSM guidelines by plotting the differences between absolute $\dot{V}O_{2\text{ PEAK}}$ measured in tests that conformed to those guidelines and those shorter or longer. Figure 3.22 shows that there was no significant difference in absolute $\dot{V}O_{2\text{ PEAK}}$.

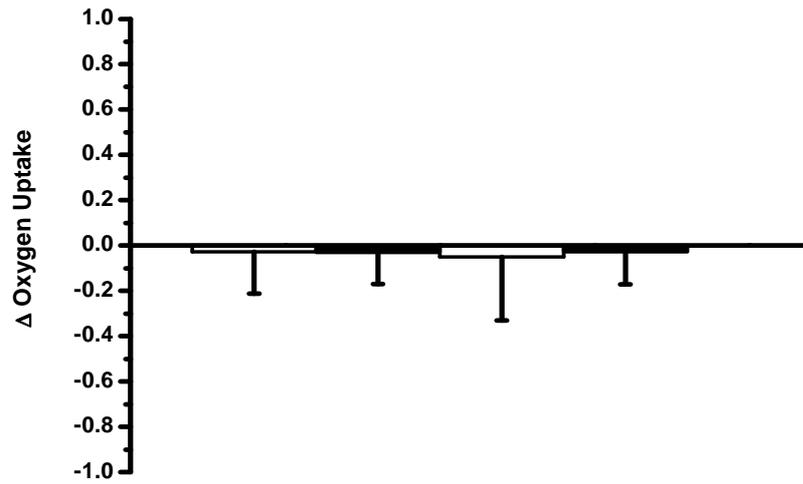


Figure 3-22 shows the effect of exercise duration not conforming to the current ACSM guidelines on absolute $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in $\text{l}\cdot\text{min}^{-1}$ and the first two columns show the difference in absolute oxygen consumption when exercise was shorter than the ACSM guidelines. The last two columns show the difference in absolute oxygen consumption when exercise was longer than the ACSM guidelines.

Similarly, figure 3.23 shows that there was no significant difference in relative

$\dot{V}_{O_2 \text{ PEAK}}$.

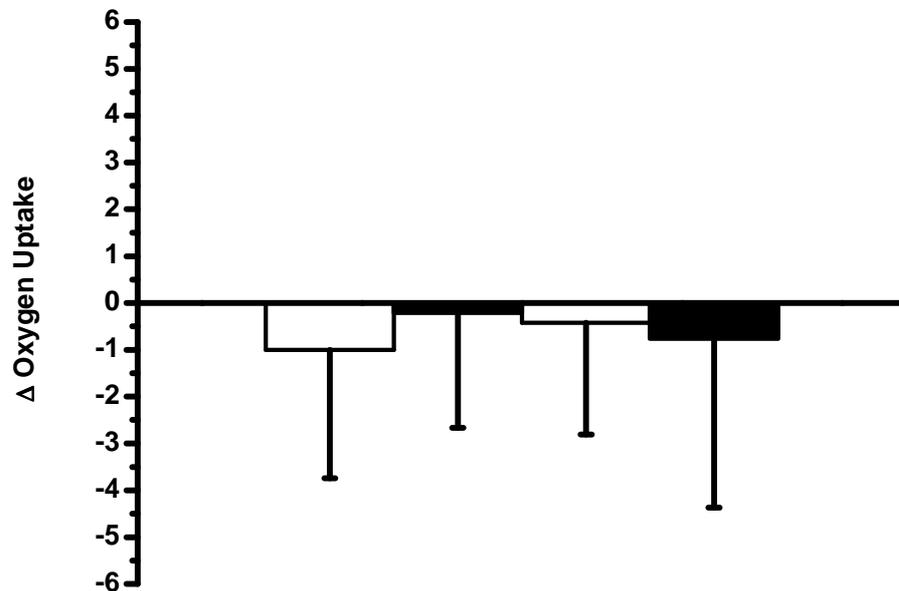


Figure 3-23 shows the effect of exercise duration not conforming to the current ACSM guidelines on relative $\dot{V}_{O_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak oxygen uptake is in ml⁻¹.kg⁻¹.min⁻¹ and the first two columns show the difference in absolute oxygen consumption when exercise was shorter than the ACSM guidelines. The last two columns show the difference in absolute oxygen consumption when exercise was longer than the ACSM guidelines.

Influence of test ramp protocols on other cycle ergometer peak parameters

Figure 3.24 shows the impact of short, moderate and long test ramp protocols (see figure 3.24 and associated text) on the $\dot{V}_{CO_2 \text{ PEAK}}$ achieved by the male and female volunteers. Males achieved $\dot{V}_{CO_2 \text{ PEAK}}$ of 3.4 ± 0.6 (range 2.4-4.4) l.min⁻¹ during the short protocol, 3.3 ± 0.5 (range 2.4-3.9) l.min⁻¹ during the moderate protocol and 3.3 ± 0.5 (range 2.5-4.2) l.min⁻¹ during the long protocol. There was no significant difference in the $\dot{V}_{CO_2 \text{ PEAK}}$ achieved by the males in the test ramp protocols. Females achieved $\dot{V}_{CO_2 \text{ PEAK}}$ of 2.3 ± 0.5 (range 1.7-3.1) l.min⁻¹ during the short protocol, 2.1 ± 0.4 (range 1.5-2.7) l.min⁻¹ during the moderate protocol and 2.1 ± 0.5 (range 1.4-2.7) l.min⁻¹ during the long protocol. Like their male

counterparts, there was no significant difference in $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ achieved by the females in the test ramp protocols.

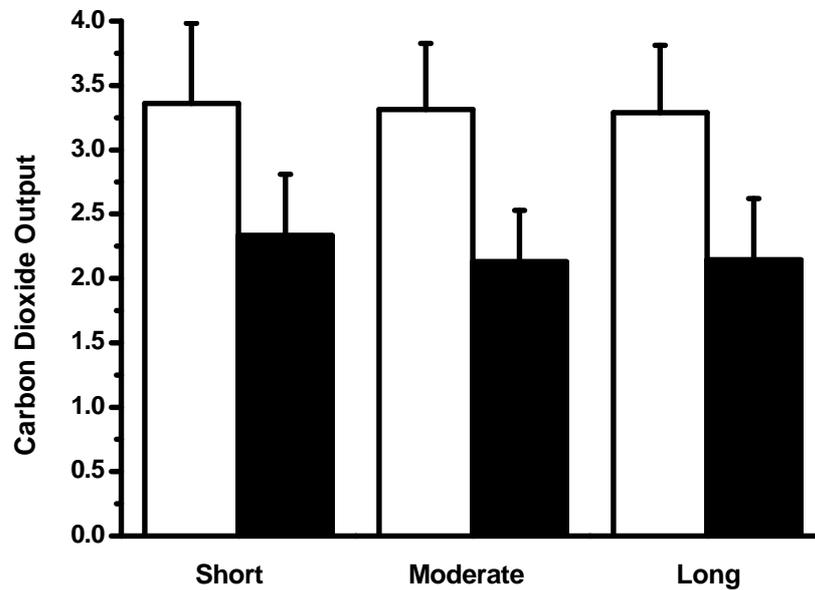


Figure 3-24 shows the effect of the test ramp cycle ergometer protocols on $\dot{V}_{\text{CO}_2 \text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak carbon dioxide output is in $\text{l}\cdot\text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Figure 3.25 shows the impact of short, moderate and long test ramp protocols (see figure 3.25 and associated text) on the $\dot{V}_{\text{E PEAK}}$ achieved by the male and female volunteers. Males achieved $\dot{V}_{\text{E PEAK}}$ of 115 ± 29 (range 75.4-166.8) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 105 ± 23 (range 69.3-146.2) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 111 ± 26 (range 61.6-147) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. There was no significant difference in the $\dot{V}_{\text{E PEAK}}$ achieved by the males in the test ramp protocols. Females achieved $\dot{V}_{\text{E PEAK}}$ of 84 ± 16 (range 58.5-105) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 82 ± 18 (range 51-113.5) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 83 ± 20 (range 50.5-113) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in $\dot{V}_{\text{E PEAK}}$ achieved by the females in test ramp protocols.

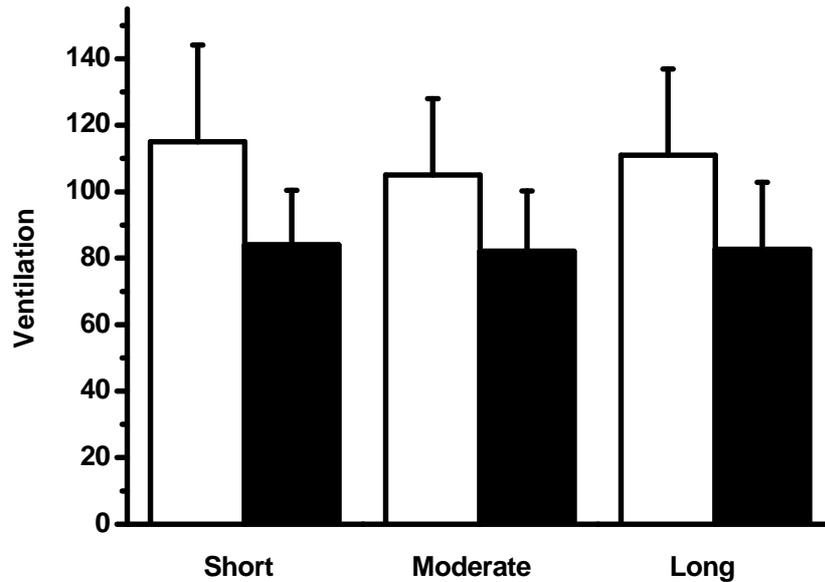


Figure 3-25 shows the effect of the test ramp cycle ergometer protocols on $\dot{V}_{E\text{ PEAK}}$. Male volunteers are represented by the open bars and female volunteers by the closed bars. Peak ventilation is in $\text{l}\cdot\text{min}^{-1}$ and the short, moderate and long test ramp protocols are described in the methods section.

Figure 3.26 shows the impact of short, moderate and long test ramp protocols (see figure 3.26 and associated text) on the $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the male and female volunteers. Males achieved a $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio of 35.3 ± 4.3 (range 29.1-41.3) during the short protocol, 32.3 ± 4.1 (range 26.5-41.0) during the moderate protocol and 34.6 ± 5.3 (range 25.0-42.3) during the long protocol. There was no significant difference in the $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the males in the test ramp protocols. Females achieved a $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio of 35.9 ± 4.5 (range 29.2-44.7) during the short protocol, 37.9 ± 6.4 (range 30.9-51.9) during the moderate protocol and 38.7 ± 5.5 (range 29.2-48.2) during the long protocol. Like their male counterparts, there was no significant difference in $\dot{V}_E / \dot{V}_{\text{CO}_2}$ ratio achieved by the females in the test ramp protocols.

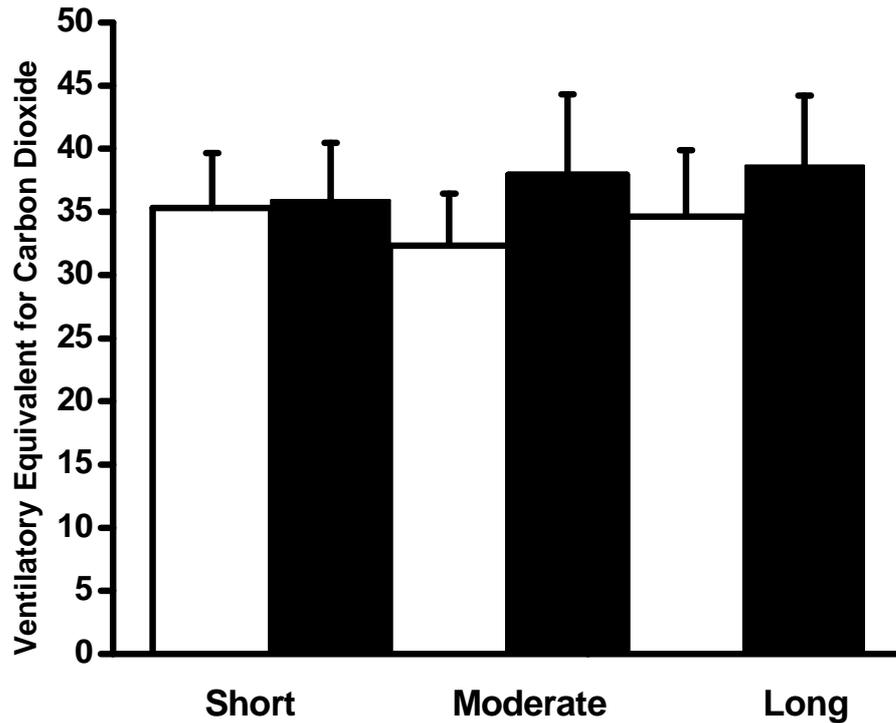


Figure 3-26 shows the effect of the test ramp cycle ergometer protocols on $\dot{V}_E / \dot{V}_{CO_2}$ ratio. Male volunteers are represented by the open bars and female volunteers by the closed bars. The short, moderate and long test ramp protocols are described in the methods section.

As stated previously, Bland-Altman plots are statistically inappropriate for testing agreement between different protocols applying the same methodology but have been applied to visualise limits of agreement and establish bias. Figure 3.27 shows that there is good agreement in various parameters across all the participants.

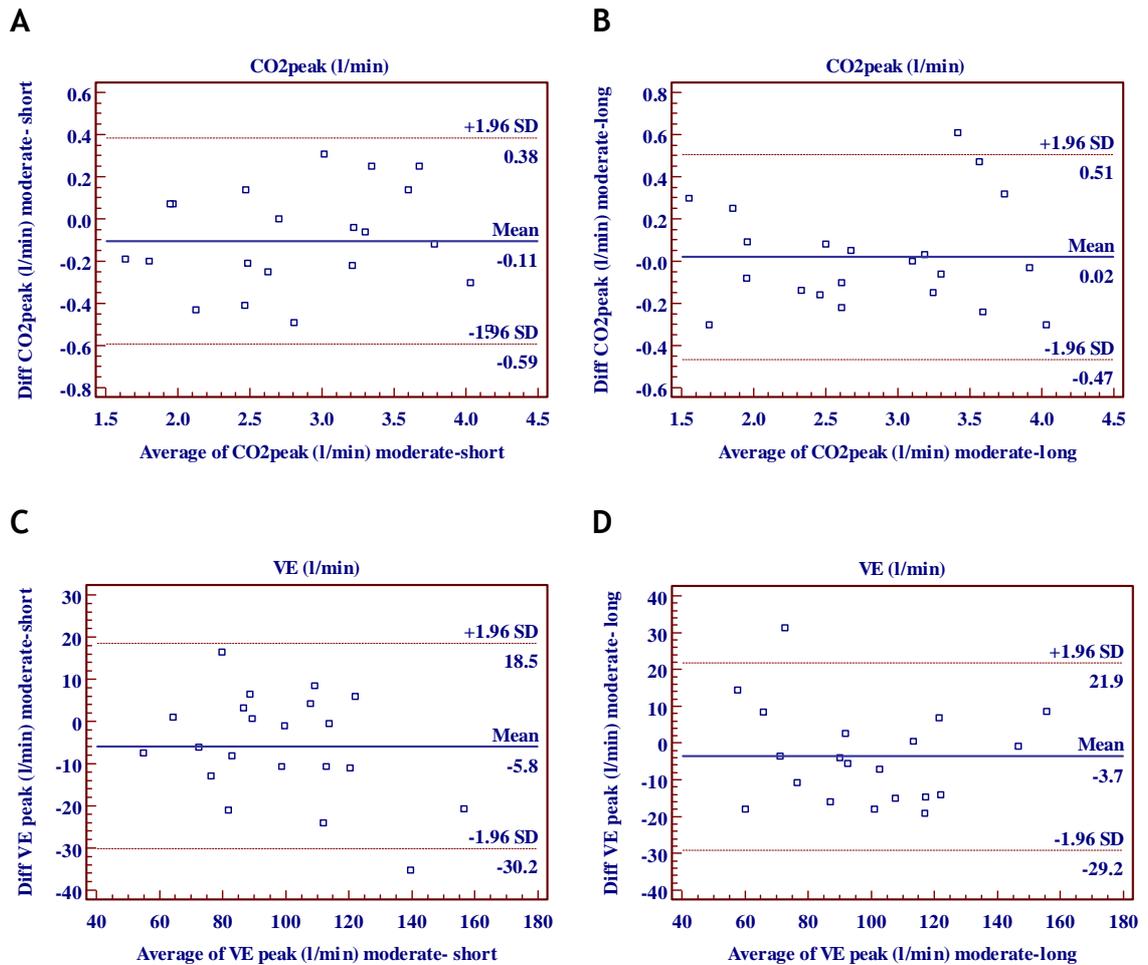


Figure 3-27 Bland-Altman plots showing agreement in $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ and \dot{V}_E measurements across all study participants. The moderate test ramp protocol was used as the standard and plotted against the short or long test ramp protocols. Panel A illustrates absolute $\dot{V}_{\text{CO}_2 \text{ PEAK}}$ with the short test ramp protocol and B the long test ramp protocol. Panel C illustrates \dot{V}_E with the short test ramp protocol and D with the long test ramp protocol.

Figure 3.28 shows the impact of short, moderate and long test ramp protocols (see figure 3.28 and associated text) on the peak RER achieved by the male and female volunteers. Males achieved a peak RER of 1.15 ± 0.05 (range 1.07-1.23) during the short protocol, 1.14 ± 0.10 (range 1.05-1.25) during the moderate protocol and 1.10 ± 0.06 (range 0.97-1.2) during the long protocol. The peak RER achieved by the males in the short test ramp protocol was significantly higher than in the long test ramp protocol ($P < 0.05$ by repeated measures). Females achieved a peak RER of 1.22 ± 0.10 (range 1.09-1.31) during the short protocol, 1.14 ± 0.10 (range 1-1.3) during the moderate protocol and 1.11 ± 0.10 (range 1-1.2) during the long protocol. There was no significant difference in peak RER achieved by the females in the test ramp protocols.

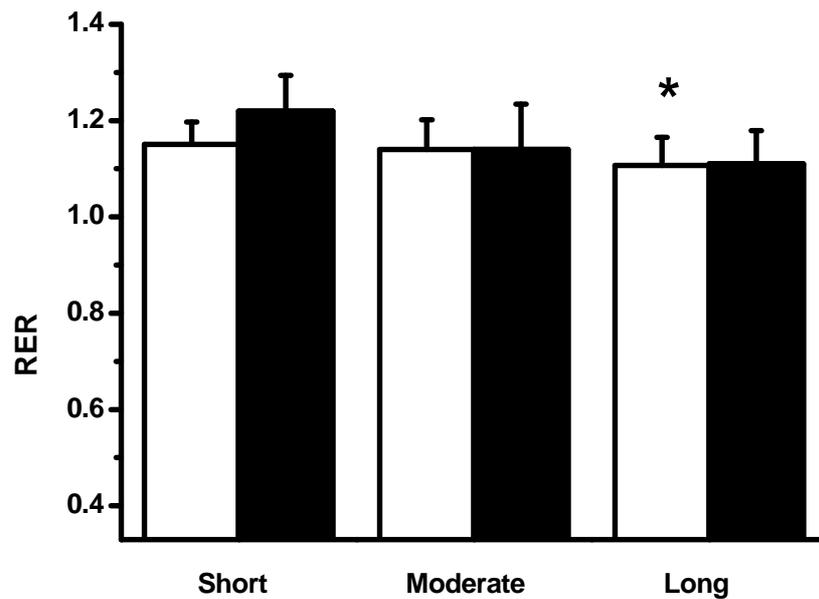


Figure 3-28 shows the effect of the test ramp cycle ergometer protocols on peak RER. Male volunteers are represented by the open bars and female volunteers by the closed bars. The short, moderate and long test ramp protocols are described in the methods section. * $P < 0.05$ compared with the short test ramp protocol in males.

Figure 3.29 shows the impact of short, moderate and long test ramp protocols (see figure 3.29 and associated text) on the peak heart rate achieved by the male and female volunteers. Males achieved a peak heart rate of 174 ± 11 (range 157-191) during the short protocol, 176 ± 10 (range 160-194) during the moderate protocol and 179 ± 12 (range 156-191) during the long protocol. There was no significant difference in the peak heart rate achieved by the males in the test ramp protocols. Females achieved a peak heart rate of 183 ± 7 (range 174-193) during the short protocol, 180 ± 15 (range 155-200) during the moderate protocol and 179 ± 15 (range 158-200) during the long protocol. Like their male counterparts, there was no significant difference in peak heart rate achieved by the females in the test ramp protocols.

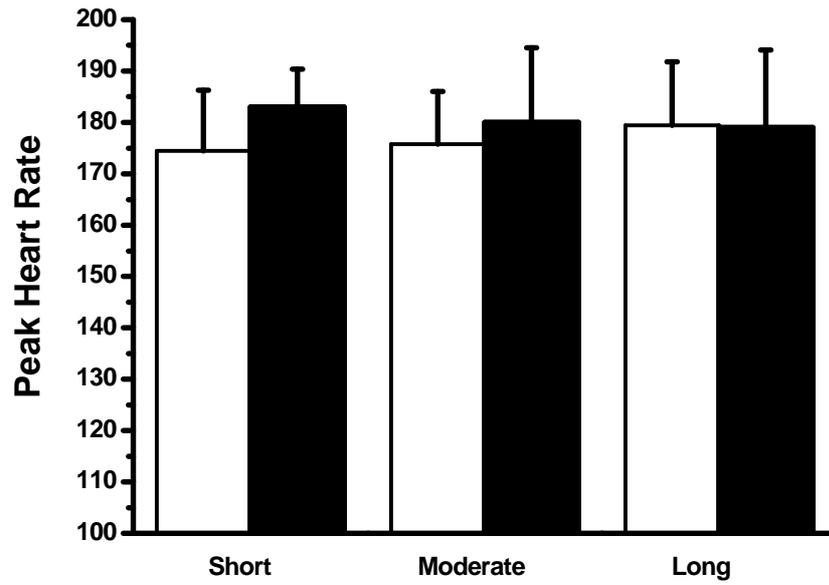


Figure 3-29 shows the effect of the test ramp cycle ergometer protocols on peak heart rate. Male volunteers are represented by the open bars and female volunteers by the closed bars. Heart rate is in beats per minute and the short, moderate and long test ramp protocols are described in the methods section.

3.7 Discussion

This study was undertaken to examine the impact of exercise duration on maximal exercise variables. This aim was achieved by re-programming the duration of individual work rate increments in a ramp Bruce protocol (Will & Walter, 1999). This approach meant that the speed/gradient increments in the treadmill tests and the resistance increments on cycle ergometer tests were constant and the only changing variable was exercise duration. Consequently, there was no significant difference in peak work loads achieved during treadmill exercise in men or women. During cycle ergometer exercise, however, the short test ramp protocol allowed men to generate higher peak work loads but no other differences were observed for men or women. Despite this higher peak work load the short test ramp protocol on the cycle ergometer had significantly shorter total exercise duration and achieved the study aim.

Thus it was possible to isolate the effects of changing exercise duration on commonly applied markers of functional capacity. The current study found that changing total exercise duration in isolation had no significant effect upon peak oxygen uptake ($\dot{V}_{O_2 \text{ PEAK}}$), peak carbon dioxide output ($\dot{V}_{CO_2 \text{ PEAK}}$), peak minute ventilation ($\dot{V}_{E \text{ PEAK}}$), the ventilatory equivalent for carbon dioxide ($\dot{V}_E / \dot{V}_{CO_2}$) and peak heart rate HR_{peak} . It was noted that the female participants had, in general, significantly lower total exercise durations than their male counterparts and that this was associated with lower measurements of $\dot{V}_{O_2 \text{ PEAK}}$, $\dot{V}_{CO_2 \text{ PEAK}}$ and $\dot{V}_{E \text{ PEAK}}$. However, the differences in body habitus between male and females is likely to account for these differences due to the higher proportions of lean body mass observed in men. For example, (Nindl *et al.*, 1998) demonstrated that $\dot{V}_{O_2 \text{ PEAK}}$ was 56% greater in men than women and (Bhambhani & Maikala, 2000) showed that men have a better \dot{V}_{O_2} response to random allocation of a weighted vest during treadmill walking.

Other studies have also found no significant difference in peak functional capacity measurements when using different exercise protocols. For example, (Kang *et al.*, 2001) found that the $\dot{V}_{O_2 \text{ PEAK}}$ in treadmill running was not significantly different when using Astrand, Bruce or Costill/Fox exercise protocols. The total exercise durations were similar to that in the current study at 9.8 minutes the

Astrand, 12.4 minutes for the Bruce and 5 minutes for the Costill/Fox (Kang *et al.*, 2001). However, the nature of the speed and gradient increments in the protocols are very different and does not reflect the sole effect of exercise duration. The measurement of $\dot{V}_{O_2 \text{ PEAK}}$ may be independent of exercise duration but may be influenced by the pattern of increased work rate.

This is illustrated by the standard Bruce protocol which has some advantages, for example, its use in many published studies provides a plethora of reference data and the three minute work load duration allows steady-state sub-maximal data to be acquired. However, the large work load increments between the three minute stages can be a disadvantage where some are forced to stop exercising prematurely because of an inability to tolerate the high work load increments. This can reduce the sensitivity of (CPET) to determine the impact of therapeutic interventions (Northridge *et al.*, 1990). Similarly, while (Bishop *et al.*, 1998) and (Zhang *et al.*, 1991) both concluded that $\dot{V}_{O_2 \text{ PEAK}}$ will be similar for all exercise durations when the pattern of work load increment is the same, they suggested that reducing the duration between work load increments provides a more precise $\dot{V}_{O_2 \text{ PEAK}}$ measurement in an exercise protocol. This concept is supported by Myers *et al* who found that $\dot{V}_{O_2 \text{ PEAK}}$ measurement is more accurate using small work load increment protocols (Myers *et al.*, 1991).

Nevertheless, Kuipers *et al* altered the work load interval in three different treadmill protocols (using stage durations of one, three and six minutes) and found similar $\dot{V}_{O_2 \text{ PEAK}}$ in eight, well motivated, endurance trained runners (Kuipers *et al.*, 2003). Thus, the level of fitness, strength and motivation to attain maximum effort in the test population may directly influence the outcome of (CPET).

Froelicher *et al* (1975) monitored $\dot{V}_{O_2 \text{ PEAK}}$ in sedentary and active participants using Balke and standard Bruce protocols. In this study, $\dot{V}_{O_2 \text{ PEAK}}$ was not different in the analysis of all sedentary participants despite significantly different total exercise durations (fifteen minutes in the Balke and nine minutes in the Bruce). However, a sub-group analysis demonstrated that men with higher lean body mass and blood haemoglobin levels were better able to tolerate the longer Balke protocol (Froelicher, Jr. *et al.*, 1974). The shorter Bruce protocol was associated

with significantly higher respiratory quotients in all participants and was similar to the findings of the current study where a lower RER was measured in the long duration test ramp protocols. This may indicate that shorter duration protocols can influence whole body metabolic response more than longer duration protocols in healthy populations. This may be more important for (CPET) in chronic medical conditions where the cardiopulmonary response to exercise can be challenged more than, for example, thermoregulation and volition to continue in extended exercise.

Respiratory exchange ratio (RER) is the ratio of carbon dioxide released to oxygen taken in, by exchange between the body and atmosphere, over a period of measurement by analysis of expired gases. In the steady state, it is equal to the respiratory quotient which reflects the proportion of different nutrients being used for energy production. When the ratio is in excess of 1.00, carbon dioxide is being released in excess of oxygen uptake and demonstrates additional non-metabolic production of carbon dioxide from bicarbonate buffering of a metabolic acidosis. This excess carbon dioxide stimulates ventilation to washout the non-metabolic carbon dioxide and consequently greater cardiopulmonary stress.

Metabolic heat production increases in proportion to the intensity of muscular activity (Saltin & Hermansen, 1966) and consequently exercise that achieves the same peak work load should have the same thermal stress. However, heat transfer from working muscle is complex and there is a temporal delay in the rise of core body temperature compared to that observed in the muscle. Longer exercise durations can facilitate heat transfer and allow a rise in core temperature that may limit exercise capacity (Gonzalez-Alonso *et al.*, 1999) without the same stress on the cardiopulmonary system. Paradoxically, the increased body temperature with longer duration exercise may elevate \dot{V}_{O_2} due to redistribution of blood flow and lead to similar $\dot{V}_{O_2\text{ PEAK}}$ measurements when compared with shorter duration protocols. Thus, it is important that $\dot{V}_{O_2\text{ PEAK}}$ is not considered in isolation when interpreting the influence of exercise duration on the validity of (CPET).

The ventilatory equivalent describes the ratio of ventilation to oxygen uptake or carbon dioxide output. For oxygen, it is an index of the efficiency of oxygen uptake that will be discussed later in this thesis. For carbon dioxide, it is an index of ventilatory drive but there is no consensus on how it should be measured. Near the end of exercise, $\dot{V}_E / \dot{V}_{CO_2}$ is non-linear because ventilation is driven both by CO₂ output and by a decrease in plasma pH (Davis *et al.*, 2006). (Milani *et al.*, 1996) found that $\dot{V}_E / \dot{V}_{CO_2}$ was higher at rest than at the peak exercise effort but attributed this to hyperventilation at the starting of exercise. So to prevent similar problems in this study, exercise was not initiated if hyperventilation was evident from monitoring the expired gases and ventilation at rest and the test ramp exercise protocol were all initiated with a short period of unloaded exercise. Furthermore, with longer work load durations, subjects might maintain high ventilations for lengthy periods leading to respiratory muscle fatigue (Whipp *et al.*, 1981; Buchfuhrer *et al.*, 1983). However, no significant difference was observed in peak $\dot{V}_E / \dot{V}_{CO_2}$ during the current study and as would be expected, there was no significant difference in its component parts \dot{V}_E or \dot{V}_{CO_2} . Tabet *et al* (2003) suggested that measuring $\dot{V}_E / \dot{V}_{CO_2}$ across the exercise duration has greater prognostic value than peak measurements (Tablet *et al.*, 2003) and emphasises that sub-maximal measurements of functional capacity are important in clinical practice.

The six minute walk test is a functional test that can be used to evaluate exercise capacity in patients with marked left ventricular dysfunction or peripheral arterial occlusive disease who cannot perform cycle ergometer or treadmill exercise. Patients are instructed to walk around a marked course at their own pace, attempting to cover as much ground as possible in six minutes. At the end of the six minute interval, the total distance walked is determined and the symptoms experienced by the patient are recorded. While this test uses a sub-maximal levels of physiological stress and only correlates modestly with $\dot{V}_{O_2 \text{ PEAK}}$, it has good prognostic power (Alameri *et al.*, 2009).

Perhaps, on the balance of conflicting evidence available for the effects of exercise duration on maximal indices of functional capacity and the functional capacity of the relevant patient populations, it might be appropriate to suggest that the current ACSM guidelines for clinical exercise testing are inappropriate.

The current study suggests that, using standardised work load increments that changing only exercise duration and in a diverse population, short duration exercise is viable to assess peak functional capacity. Even when data was categorised to fit exercise durations consistent with the Buchfuhrer study (Buchfuhrer *et al.*, 1983) it was apparent that there was no difference in the vast majority of the outcome measures.

Shorter total exercise duration protocols appear to stress the cardiopulmonary system appropriately to allow peak responses to be measured in both active and sedentary healthy populations (Kuipers *et al.*, 2003; Froelicher, Jr. *et al.*, 1974). However, even in this study of healthy volunteers the females generated less peak work loads than the men and consequently special attention needs to be paid to 'frail' populations where they cannot generate true maximal efforts because of strength limitations.

This gender bias can be extrapolated to patient populations where their clinical condition may prevent or make maximal exercise testing inappropriate. Under such circumstances, short duration tests may be appropriate for generating peak data but could limit their application for sub-maximal data analysis. That is to say that a short duration protocol might provide sparse data around metabolically relevant points in breath by data rendering them inappropriate and enhancing the validity of current ACSM guidelines. It is, therefore, important to also establish the effects of exercise duration on sub-maximal markers of functional capacity to ensure that evidence based practice is in place for (CPET) in clinical populations.

CHAPTER FOUR

Ventilatory Anaerobic Threshold

4 Impact of Exercise Duration on Anaerobic Threshold Measurements

4.1 Introduction

Measurement of peak aerobic capacity ($\dot{V}_{O_2 \text{ PEAK}}$) can be difficult in certain patient populations due to the physiological stresses associated with maximal testing. In frail populations, sub-maximal testing may be preferred and provide a more reproducible test with better patient compliance. Typically, ventilatory anaerobic threshold VAT is one of the most common sub-maximal measurements recorded during the cardiopulmonary exercise testing (CPET). This marker can be used to assess the level of impairment and disability of subjects by measuring the level of anaerobic acidosis break point, and how early it occurs, during (CPET). It is used to determine the cardiopulmonary reserve available to perform work for at least 8 hours (Hansen *et al.*, 1984). In addition, it has been used to assess functional capacity in sporting and healthy populations (Davis *et al.*, 1976; Davis *et al.*, 1979; Davis, 1985).

Ventilatory anaerobic threshold has been found to be a good estimate of functional capacity in chronic heart failure patients (Lipkin *et al.*, 1985). It can be measured during a sub-maximal test and can distinguish hyperventilation coming from stress or anxiety with hyperventilation coming from hypoxia (due to the lack of an adequate oxygen supply). The technique has also been used to assess pre-operative risk in the elderly (Sinclair *et al.*, 2009) and high risk patients undergoing surgery for abdominal aortic aneurysms (Kothmann *et al.*, 2009).

The direct measurement of anaerobic threshold during (CPET) requires the use of mildly invasive techniques that are not available in all clinics. The procedures involved in the direct measurement can take a long time to deliver results, will need specialist training in collecting venous or capillary blood samples and will require good patient co-operation. These limitations make the non-invasive

estimation of anaerobic threshold from expired gas analysis very attractive (Aunola & Rusko, 1986; Beaver *et al.*, 1985) .

Davis *et al* found that measuring an increase in expired CO₂ concentration relative to expired O₂ concentration was strongly correlated with an increase in blood lactate concentration during (CPET) (Davis *et al.*, 1976). Further studies have reported that VAT is related to the point where energy switches from aerobic to anaerobic sources during exercise (Yoshida *et al.*, 1981; Aunola & Rusko, 1986). The term ventilation at anaerobic threshold VAT refers to the onset of an exercise induced hyperventilation during (CPET) which sequentially increase the volume of minute ventilation (\dot{V}_E) to remove the excess CO₂ produced by bicarbonate buffering of H⁺ from lactic acid (Wasserman & Mcilroy, 1964; Wasserman & Whipp, 1975).

The actual determination of VAT appears relatively easy and is identified as a non-linear increase ventilation with increasing work rate. This is often presented in clinical situations as a plot of \dot{V}_{O_2} (to represent increasing work rate) against \dot{V}_{CO_2} (to represent the stimulus for ventilation) and is called the V-slope (Wasserman & Mcilroy, 1964; Wasserman *et al.*, 1973). However, the exact point where an increase in ventilation occurs can often be difficult to distinguish. Therefore, further procedures have been suggested to accurately define anaerobic threshold break point by using two additional standard gas exchange criteria ($\dot{V}_E / \dot{V}_{O_2}$ and $\dot{V}_E / \dot{V}_{CO_2}$) against \dot{V}_{O_2} .

A detailed discussion of these procedures has been presented previously, but in summary the accuracy in determining the VAT break point is significantly improved in research laboratories by using two additional standard gas exchange criteria. Specifically, the fraction of end tidal CO₂ (F_{ET}CO₂%) and the ventilatory equivalents for oxygen and carbon dioxide ($\dot{V}_E / \dot{V}_{O_2}$ and $\dot{V}_E / \dot{V}_{CO_2}$) can be plotted against \dot{V}_{O_2} with data deleted after the respiratory compensation point RCP and isocapnic buffering period (Whipp *et al.*, 1989; Wasserman *et al.*, 1967; Wasserman *et al.*, 1973; Wasserman *et al.*, 1977; Myers & Ashley, 1997; Caiozzo *et al.*, 1982; Bischoff & Duffin, 1995; Beaver *et al.*, 1986; Davis *et al.*, 1976; Scheuermann & Kowalchuk, 1999).

In the clinical environment, the V-slope method is generally used exclusively to determine the VAT break point because of the time constraints in data analysis. Most automated gas analysis equipment will use algorithms that employ the V-slope technique to generate their test reports and a recent study acknowledges that it can be difficult to establish the VAT break point in the breath-by-breath data from frail patients (Hopker *et al.*, 2011).

While the precise factors that make it difficult to establish the VAT break point remains unclear it is generally the case that most tests are conducted under the current ACSM guidelines for clinical exercise testing. As these guidelines aim to reach maximum effort within eight to twelve minutes it is obvious that any sub-maximal effort could produce even shorter exercise durations. Short exercise durations might compromise the accuracy of linear fitting and subsequent break point determination by limiting the volume of breath-by-breath data recorded during exercise around the anaerobic threshold.

The adverse effect of short exercise duration on the quantity of data available for statistical analysis will be amplified by the so-called cardio-dynamic phase during exercise. The cardio-dynamic phase, or phase I, is an abrupt hyperpnoea at onset of exercise that is not related to metabolic stimulus of ventilatory chemoreceptors (Wasserman *et al.*, 1974). The stimulus for this excessive ventilation could be related to a neurological stimulus from active muscle that occurs before an increase in \dot{V}_{CO_2} (Tibes, 1977). Phase I can influence gas exchange kinetics for up to two minutes at the beginning of exercise and can disrupt the normal linear relationship between \dot{V}_{O_2} and \dot{V}_{CO_2} . While this effect can be diluted by including an unloaded stage in the test protocol, it is often recommended that the initial period of exercise is excluded from the data analysis by data editing. Consequently, the adverse effect is more often observed in automated gas analysis systems commonly employed in clinical research laboratories due to a lack of data editing and an unloaded stage.

The aim of this study is, therefore, to determine the influence of exercise duration on the reliability of VAT break point determination.

4.2 Methods

4.2.1 Subjects

The main aim of this study is to address the issue of exercise duration and therefore the analysis has been restricted to those volunteers that managed to complete at least three test ramp protocols on either the treadmill or cycle ergometer. Some of the volunteers managed to complete the necessary test ramp protocols on both modalities while others only managed this for one. The physical characteristics for the two groups are shown in table 4.1.

Variable	Gender	Treadmill	Cycle Ergometer
Number of Participants	Male	11	11
	Female	9	9
Age (years)	Male	31.2±6.0 (24-42)	32.5±6.0 (25-42)
	Female	36.6±11.0 * (24-57)	35.4±10.0 * (24-57)
Weight (kg)	Male	76.0±7.7 (60-84)	79.0±12.5 (60-103)
	Female	60.7±9.0 * (48-77)	60.0±9.0 * (48-77)
Height (cm)	Male	175.0±6.4 (163-182)	175.4±5.7 (163-182)
	Female	162.0±9.0 * (146-175)	160.7±7.3 * (145-172)
BSA [†] (m) ²	Male	1.92±0.14 (1.94-2.02)	1.96±0.12 (1.65-2.22)
	Female	1.65±0.14 * (1.47-1.87)	1.64±0.13 * (1.47-1.87)
BMI [#] (kg.m ²)	Male	24.8±2.0 (21-28)	26.0±4.0 (21-34)
	Female	23.1±3.8 * (18-29)	23.0±4.0 * (18-29)

Table 4.1 Physical characteristics of all study volunteers. Data are presented as mean±SD (where appropriate) and the range is given in parenthesis. * indicates $P<0.05$ compared with males (t -test with Bonferroni correction). [†] BSA is Body Surface Area according to Mosteller formula. [#] BMI is Body Mass Index.

4.3 Measuring the Ventilatory Anaerobic Threshold

The non-linear break point in the V-slope of breath-by-breath data was used to determine the VAT. Figure 4.1 illustrates that it can be difficult to identify the break point and the benefits that can be derived from editing the data to exclude the cardio-dynamic phase and noisy data. The cardio-dynamic phase was excluded by removing the first two minutes of the test ramp protocol data while noisy data was removed using the process described in the previous chapter (see figure 3.1 and associated text).

The slope of the linear regressions applied to the raw and edited data were compared at short, moderate and long exercise durations (see table 4.2 and associated text) to help establish the validity of the non-linear break point determined by that data. In addition, the absolute \dot{V}_{O_2} value at the break point was compared in the test protocols to establish any difference in a functionally relevant measure of the anaerobic threshold. The 'gold standard' method of establishing the anaerobic threshold employs a combination of the V-slope with standard gas exchange criteria (*i.e.* $F_{ET}O_2\%$, $\dot{V}_E / \dot{V}_{O_2}$ and $\dot{V}_E / \dot{V}_{CO_2}$). This method was used to provide the best prediction of VAT in the short, moderate and long exercise durations and used to establish whether exercise duration *per se* influences VAT.

4.4 Statistical Analysis

The present study investigated the impact of exercise duration on VAT, \dot{V}_E , work load by using repeated measure ANOVA. All data are presented as mean \pm standard deviation (SD) and statistical analyses were performed using Origin Pro 8 SR4 v8. 0951(B951) and SPSS (18.0.0. Chicago) for each case. At the 0.05 level, all sub-maximal variables at anaerobic threshold point are normally distributed by Shapiro-Wilks normality test and suitable for parametric data analysis. All sub-maximal variables were significantly drawn from a normal distributed population. A repeated measure ANOVA was used to examine differences between VAT. If a significant was obtained, Bonferroni adjustments post hoc test was used for multiple comparisons between means The nonlinearity of regression equation of $\dot{V}_{O_2} / \dot{V}_{CO_2}$ or slope (V-slope) was used determined the anaerobic

threshold break point and the values of VAT were compared with standard gas exchange criteria (i.e. $F_{ET}O_2\%$, $\dot{V}_E / \dot{V}_{O_2}$ and $\dot{V}_E / \dot{V}_{CO_2}$) values. Statistical significance was analysed by using Two-Sample t-test to compare between the physical characteristic of two modalities.

Moreover, linear regression were derived in order to describe the goodness of fit of a model (giving an adjusted R^2 value) of the breath-by-breath data before and after the VAT. The adjusted R^2 value represents the percentage of vertical variance from a line fitted to a simple linear regression analysis. In regression, the adjusted R^2 coefficient of determination is a statistical measure of how well the regression line approximates the real data points. And An adjusted R^2 of 1.0 indicates that the regression line perfectly fits the data.

For comparison of VAT from Raw and editing data, a paired Student's t-test was used. And P -values <0.05 were considered significant.

A simple linear regression analysis can be used to examine the goodness of fit (giving an adjusted R^2 value) of the breath-by-breath data before and after the VAT. Furthermore, outlying data that reduces the precision of the linear regression of V-slope identified by a residual sum of square RSS analysis. Which is represents unexplained (or residual) variation after fitting a regression model

4.5 Results

4.5.1 Physical characteristics

Table 4.1 indicates that the study participants are heterogeneous showing a wide range in age and body habitus. However, there was no significant difference in the physical characteristics between the two exercise modality groups. As expected, the male volunteers were significantly taller and heavier (resulting in higher BSA and BMI) than the female volunteers. The female group were significantly older than the males.

4.6 Influence of test ramp protocols on exercise duration

The different biomechanics and energy demands of walking and cycling can influence cardiopulmonary measurements during sub-maximal effort. Consequently, as with the previous chapter, the data for each exercise modality are presented separately.

Influence of exercise duration on linear regression to determine break point

The break point of the V-slope can be identified by plotting straight lines on the plot of \dot{V}_{O_2} against \dot{V}_{CO_2} . One line is plotted to the data above the VAT and the other below. The relationship is steeper above the VAT due to metabolic production of CO_2 and so the point where the lines converge will show the break point. Obviously, goodness of fit may be influenced by both the quality and quantity of the data plotted. The effect of exercise duration on the fit is described here.

Exercise duration	Treadmill			
	Before AT		After AT	
	Male	Female	Male	Female
Short	0.8±0.07	0.79±0.09	1.5±0.15	1.58±0.27
Total	0.8±0.08		1.54±0.21	
Moderate	0.84±0.09	0.77±0.11	1.4±0.15	1.4±0.3
Total	0.81±0.1		1.4±0.22	
Long	0.85±0.04	0.76±0.11	1.37±0.17	1.32±0.16
Total	0.81±0.09		1.35±0.16	

Table 4.2 indicates the value of the slope in the $\dot{V}_{\text{CO}_2}/\dot{V}_{\text{O}_2}$ relationship before and after the VAT in raw data before any editing procedures.

Statistically, a simple linear regression analysis can be used to examine the goodness of fit (giving an adjusted R^2 value) of the breath-by-breath data before and after the VAT. The adjusted R^2 value represents the percentage of vertical variance from a line fitted to a simple linear regression analysis (see figure 4.1).

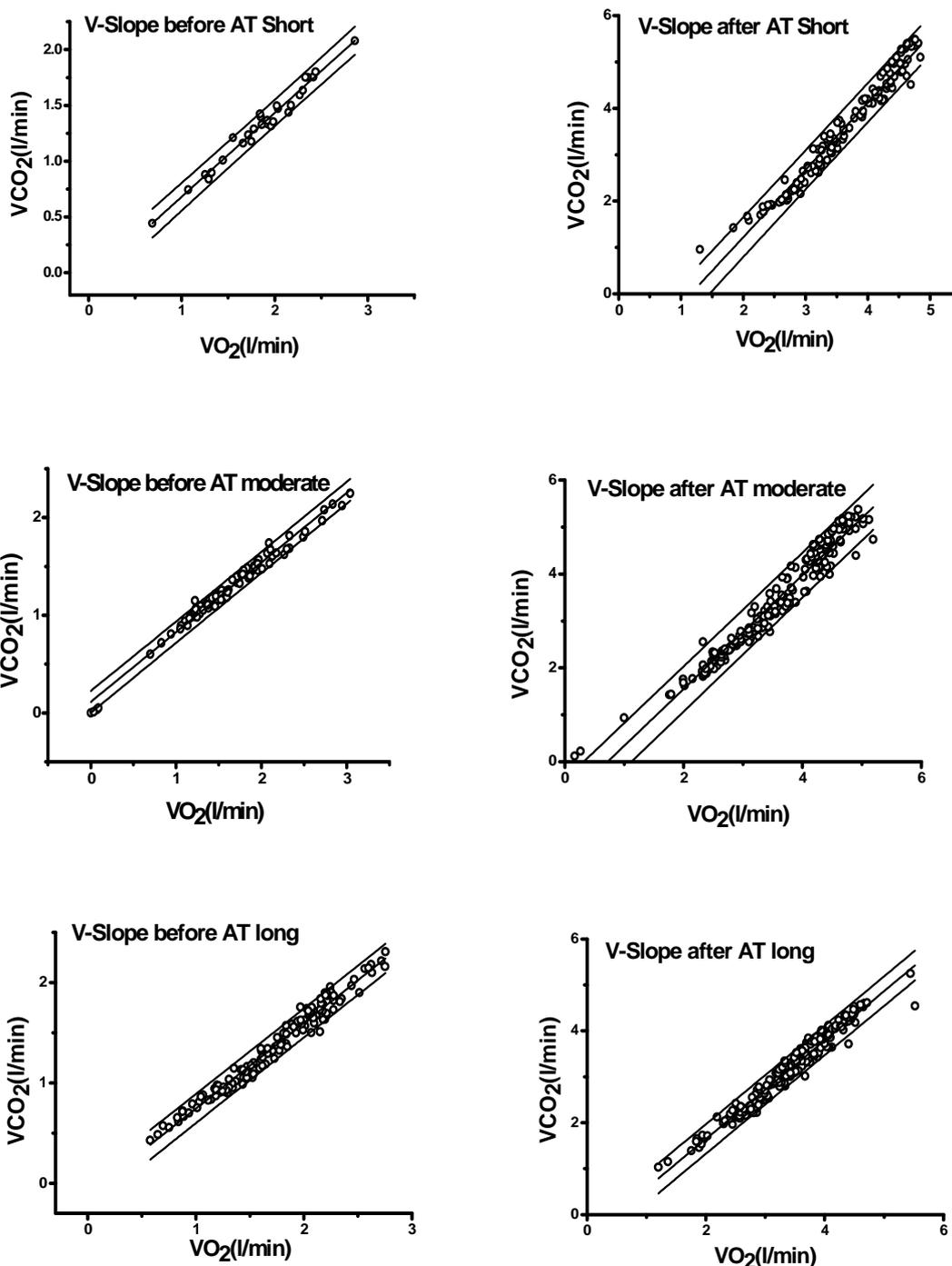


Figure 4-1 example subject (GB) of V-Slope before and after anaerobic threshold showing how goodness (Adjust R^2) and of breath by breath around the regression line

Figure 4.2 illustrates that the adjusted R^2 values are higher before the VAT and that the values increase in proportion to exercise duration. In this example, the data was edited by removing the cardio-dynamic phase and noisy data prior to then linear regression. The adjusted R^2 values for all subjects were 0.97 ± 0.02 before the VAT and 0.92 ± 0.05 after the VAT during the short protocol, 0.97 ± 0.01

before the VAT and 0.94 ± 0.04 after the VAT during the moderate protocol and 0.97 ± 0.02 before the VAT and 0.96 ± 0.03 after the VAT during the long protocol. The R^2 values calculated during the short protocol were significantly lower than in the moderate, while those measured during long duration exercise were significantly higher.

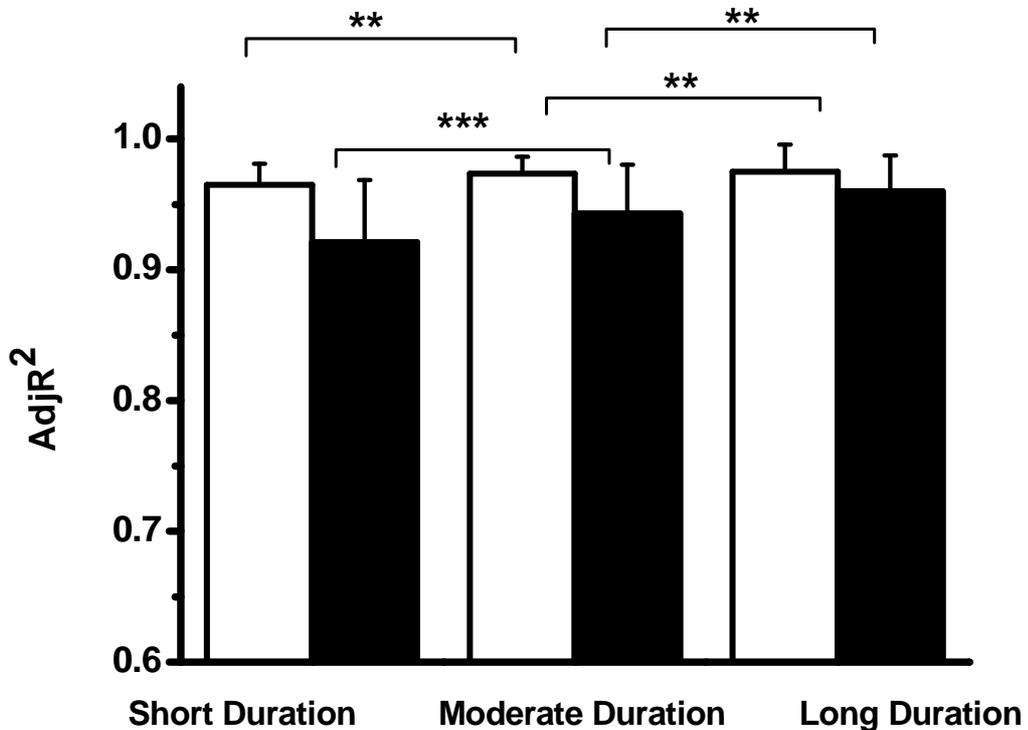


Figure 4-2 AdjR² = Adjust R² of breath by breath for V-slope before open bars and after closed bars of AT in treadmill, ** p<0.001, *** p= 0.00 significant difference from moderate duration.

Furthermore, outlying data that reduces the precision of the linear regression model can be identified by a residual sum of square RSS analysis. Figure 4.3 shows the RSS values for all subjects were 0.09 ± 0.10 before the VAT and 7.11 ± 60 after the VAT during the short protocol, 0.22 ± 0.23 before the VAT and 5.40 ± 4.80 after the VAT during the moderate protocol and 0.23 ± 0.19 before the VAT and 4.32 ± 2.20 after the VAT during the long protocol. There was no systematic difference across the test before anaerobic threshold (figure 4.3 A), however the values decrease in proportion to exercise duration (figure 4.3 B).

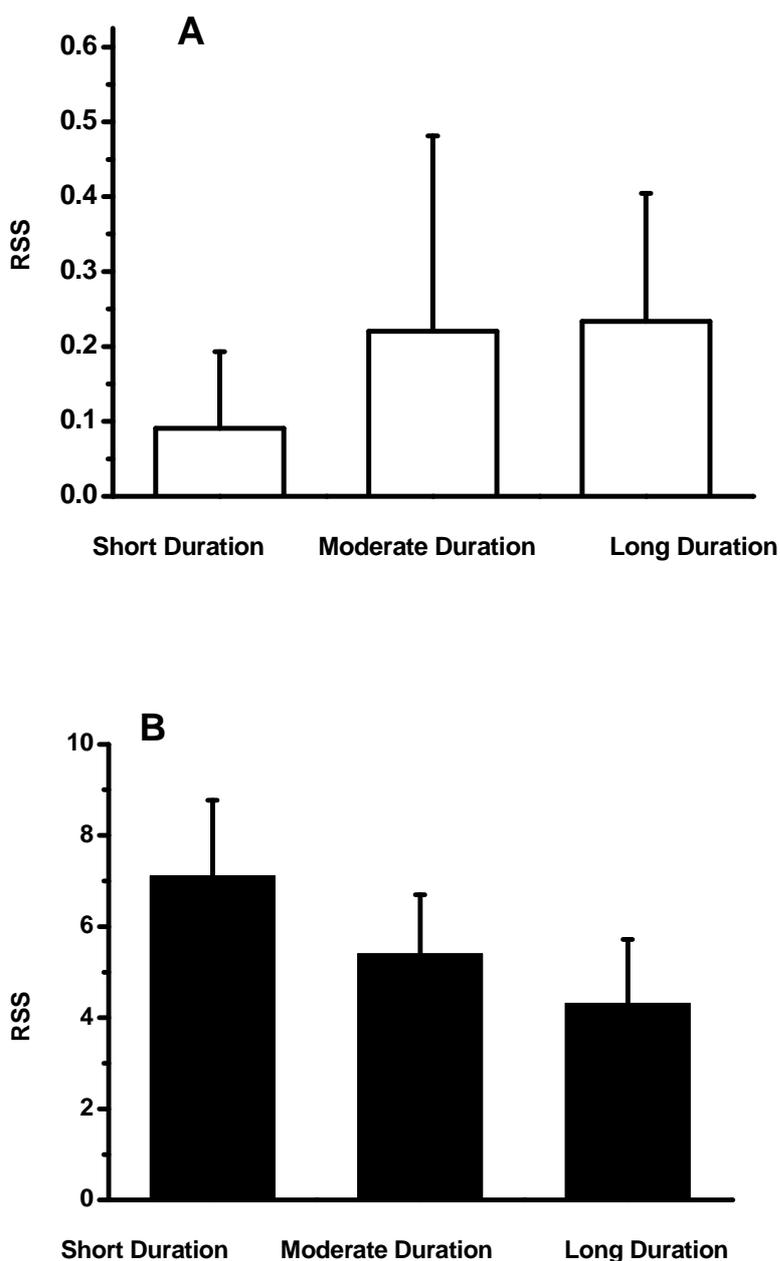


Figure 4-3 showed Residual sum of square RSS or residual (or error) of V-slope before (A open bars)and after (B closed bars)anaerobic threshold break point on treadmill, and the RSS of V-slope before AT was lower than RSS of V-slope after anaerobic threshold .

A similar relationship was shown for raw data where exercise duration influenced the goodness of fit on the linear regression analysis (data not shown in the interest of brevity). However, the physiological impact of this statistical anomaly can be assessed by determining the influence of exercise duration on the \dot{V}_{O_2} , work load and \dot{V}_E at VAT for both edited and unedited data.

Influence of test ramp protocols on treadmill VAT - with no data editing

Figure 4.4 illustrates the impact of short, moderate and long test ramp protocols (see figure 4.4 and associated text) on the VAT for male and female volunteers.

Figure 4.4 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.15 ± 0.45 (range 1.46-2.74) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.08 ± 0.25 (range 1.78-2.61) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 2.07 ± 0.37 (range 1.46-2.84) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.49 ± 0.44 (range 1.04-2.19) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.46 ± 0.40 (range 0.93-1.97) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.51 ± 0.41 (range 0.97-2.01) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

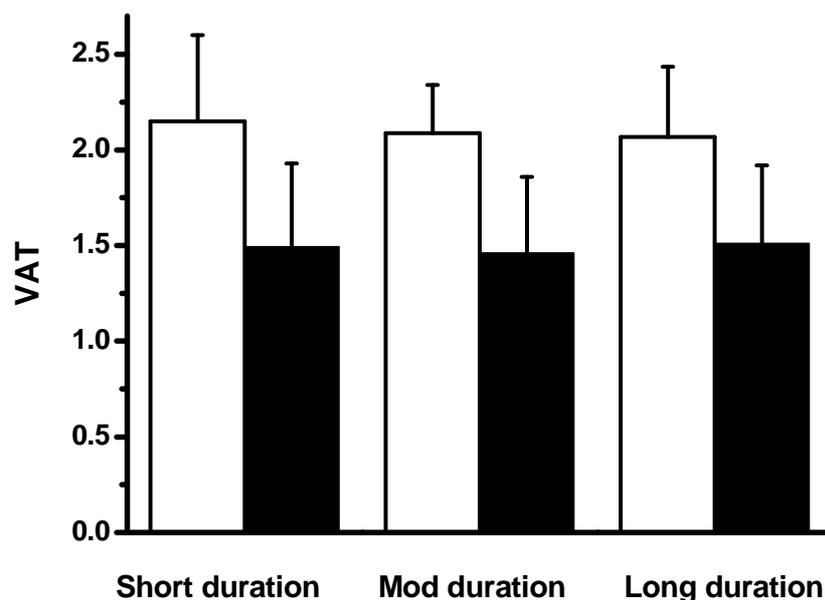


Figure 4-4 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Influence of test ramp protocols on treadmill Work - with no data editing

Similarly, figure 4.5 shows that VAT was identified in male volunteers at a work load of 128 ± 18 (range 103-164) W during the short protocol, 115 ± 19 (range 73.70-145) W during the moderate protocol and 105 ± 16 (range 88.20-135) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 100 ± 18 (range 75.20-120) W during the short protocol, 90 ± 28 (range 59.70-124) W during the moderate protocol and 76 ± 19 (range 59.60-104) W during the long protocol. Like their male counterparts, there was no significant difference in the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

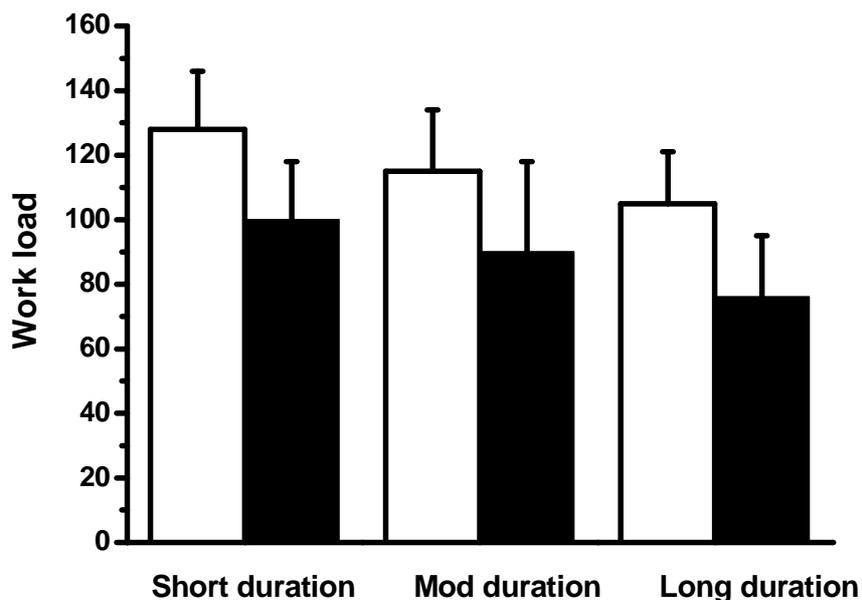


Figure 4-5 illustrates the impact of short, moderate and long test ramp protocols on work load during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Influence of test ramp protocols on treadmill \dot{V}_E - with no data editing

Furthermore, figure 4.6 shows that VAT was identified in male volunteers at \dot{V}_E of 43.30 ± 8 (range 35-55) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 43.50 ± 7 (range 32-57) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 41.70 ± 6 (range 34-51) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at \dot{V}_E of 33.60 ± 9 (range 20-43) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 33.60 ± 9 (range 23-51) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 32.10 ± 10 (range 20-49) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

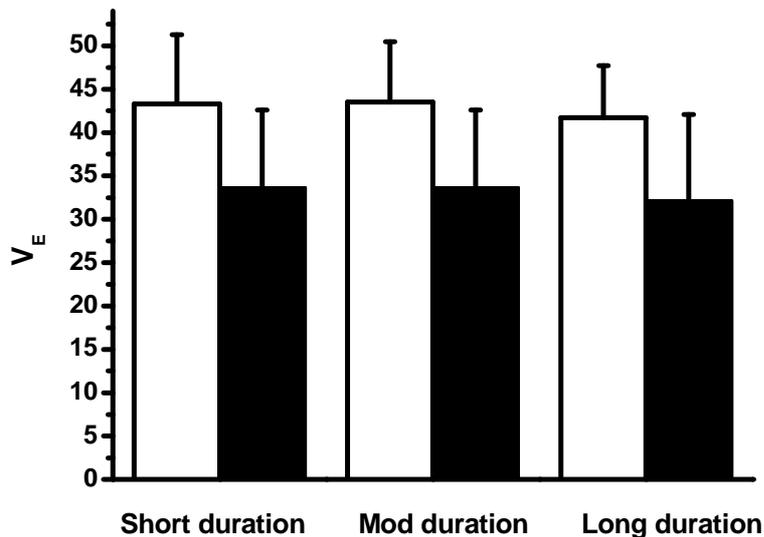


Figure 4-6 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Influence of test ramp protocols on treadmill VAT - with data editing

Figure 4.7 illustrates the impact of short, moderate and long test ramp protocols (see figure 2.1 and associated text) on the VAT for male and female volunteers.

Figure 4.7 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.01 ± 0.40 (range 1.52-2.60) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.10 ± 0.30 (range 1.80-2.60) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.96 ± 0.40 (range 1.60-2.80) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.48 ± 0.33 (range 1.14-2.06) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.55 ± 0.50 (range 1.00-2.36) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.53 ± 0.45 (range 1.00-2.30) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

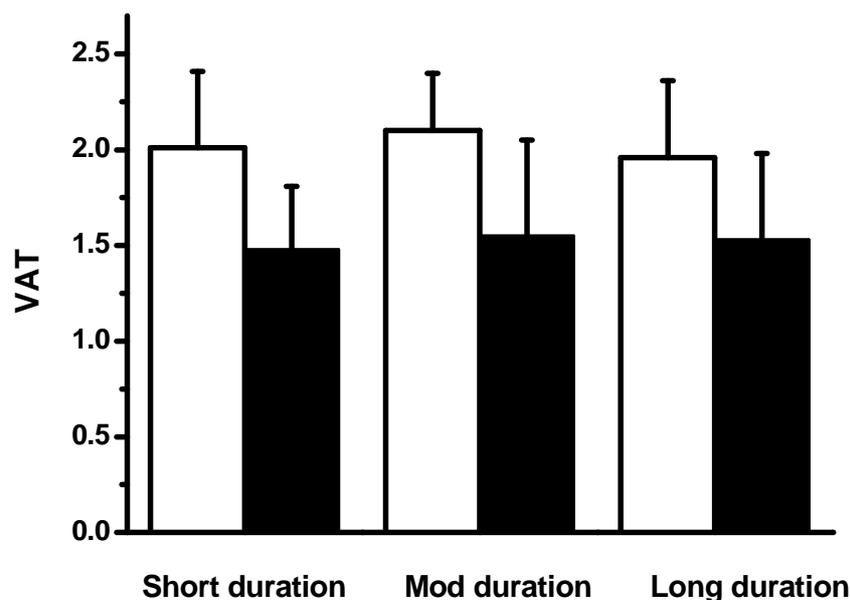


Figure 4-7 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Influence of test ramp protocols on treadmill work - with data editing

Similarly, figure 4.8 shows that VAT was identified in male volunteers at a work load of 128 ± 18 (range 103-164) W during the short protocol, 115 ± 19 (range 73.70-

145) W during the moderate protocol and 105 ± 16 (range 88.20-135) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 100 ± 18 (range 75.20-120) W during the short protocol, 90 ± 28 (range 59.70-124) W during the moderate protocol and 76 ± 19 (range 59.60-104) W during the long protocol. Like their male counterparts, there was no significant difference in the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

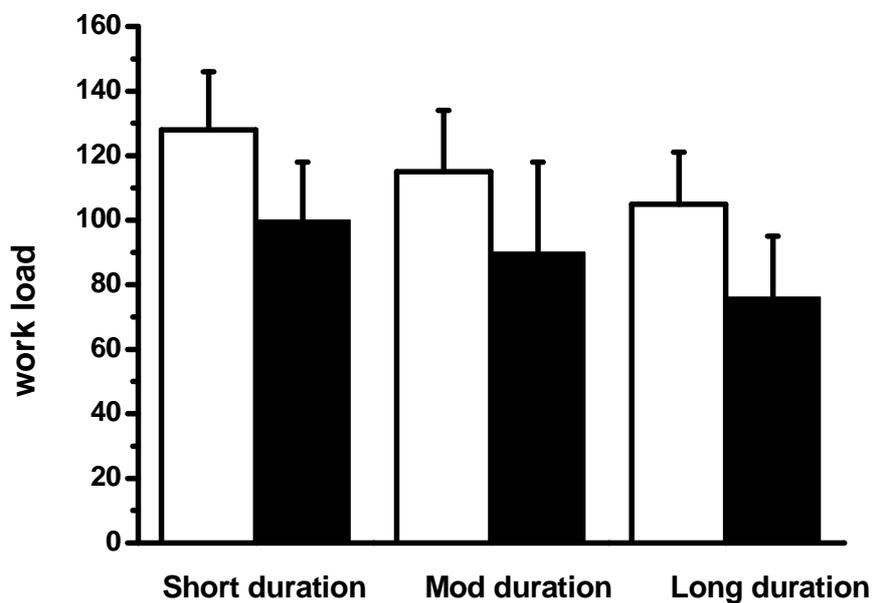


Figure 4-8 illustrates the impact of short, moderate and long test ramp protocols on the work load during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Influence of test ramp protocols on treadmill \dot{V}_E - with data editing

Furthermore, figure 4.9 shows that VAT was identified in male volunteers at \dot{V}_E of 42.60 ± 10 (range 32-65.80) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 45 ± 5.10 (range 35-54) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 41.40 ± 6 (range 32-53) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at

\dot{V}_E of 30.10 ± 6 (range 20.20-38) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 33.70 ± 9 (range 22-50) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 32.70 ± 9 (range 22-47) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

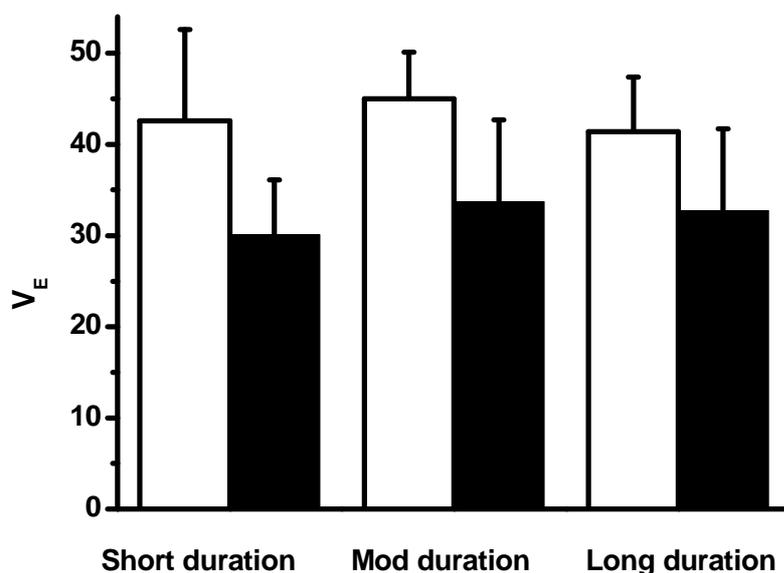


Figure 4-9 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Influence of test ramp protocols on treadmill VAT - with gas exchange data.

The use of gas exchange data allows the VAT to be identified without relying on the simple V-slope method and is therefore less sensitive to factors influencing linear regression analysis. For this reason, the influence of exercise duration can be tested on the \dot{V}_{O_2} , work load and \dot{V}_E at VAT directly rather than on artefacts of analysis.

Figure 4.10 illustrates the impact of short, moderate and long test ramp protocols (see figure 4.10 and associated text) on the VAT for male and female volunteers.

VAT by $\dot{V}_E / \dot{V}_{O_2}$ from Raw data.

Figure 4.10 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.30 ± 0.40 (range 1.62-2.76) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.20 ± 0.40 (range 1.77-3.12) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 2.20 ± 0.40 (range 1.47-2.83) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.54 ± 0.41 (range 1.13-2.22) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.56 ± 0.42 (range 0.98-2.18) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.60 ± 0.45 (range 1.09-2.32) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

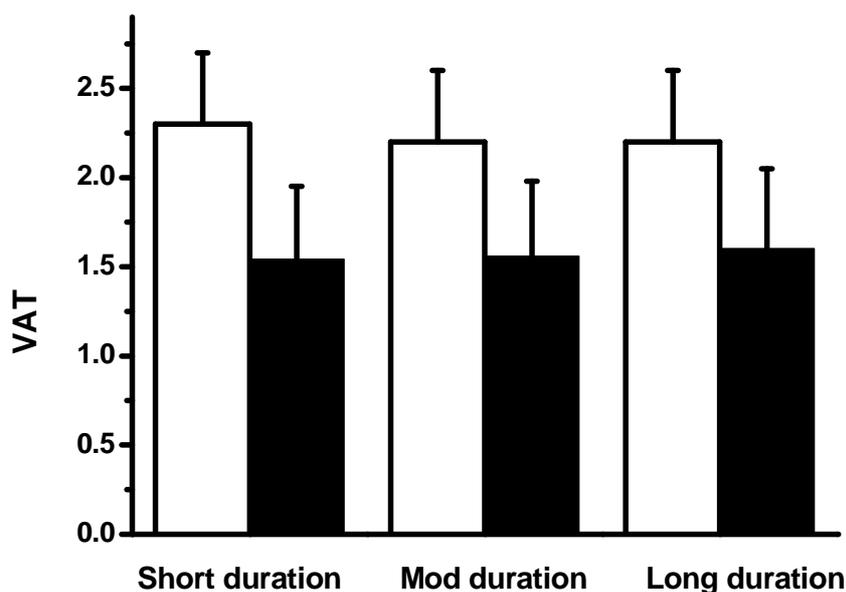


Figure 4-10 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data by $\dot{V}_E / \dot{V}_{O_2}$ method.

VAT $F_{ET}O_2\%$ from Raw data.

Figure 4.11 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.23 ± 0.40 (range 1.58-2.84) $l \cdot \text{min}^{-1}$ during the short protocol, 2.18 ± 0.40 (range 1.74-3.13) $l \cdot \text{min}^{-1}$ during the moderate protocol and 2.14 ± 0.40 (range 1.44-2.75) $l \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.53 ± 0.40 (range 1.17-2.17) $l \cdot \text{min}^{-1}$ during the short protocol, 1.54 ± 0.40 (range 0.99-2.00) $l \cdot \text{min}^{-1}$ during the moderate protocol and 1.61 ± 0.50 (range 1.03-2.31) $l \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

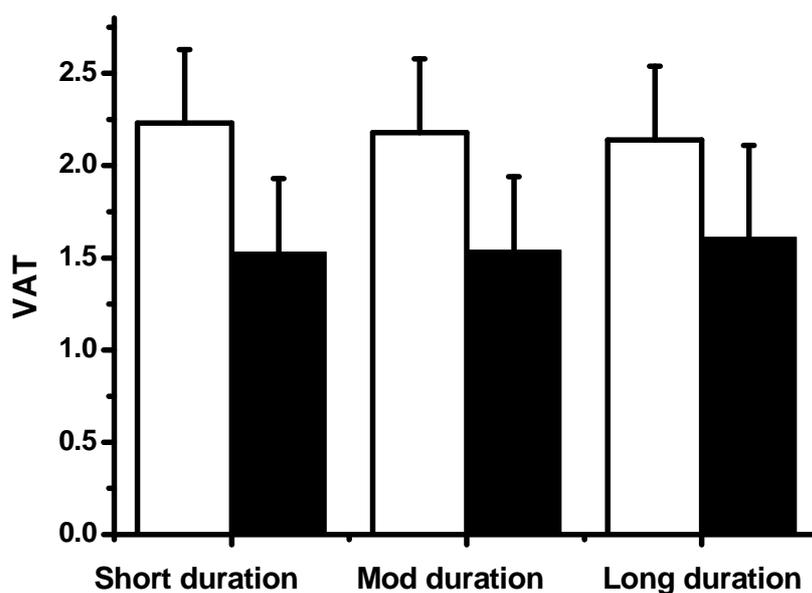


Figure 4-11 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data by $F_{ET}O_2\% / \dot{V}_{O_2}$ method.

VAT $\dot{V}_E / \dot{V}_{O_2}$ from Edit data.

Figure 4.12 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.05 ± 0.40 (range 1.55-2.67) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 2.10 ± 0.30 (range 1.79-2.62) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 2.05 ± 0.40 (range 1.63-2.85) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.55 ± 0.40 (range 1.15-2.38) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.56 ± 0.50 (range 1.01-2.36) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.58 ± 0.50 (range 1.03-2.30) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

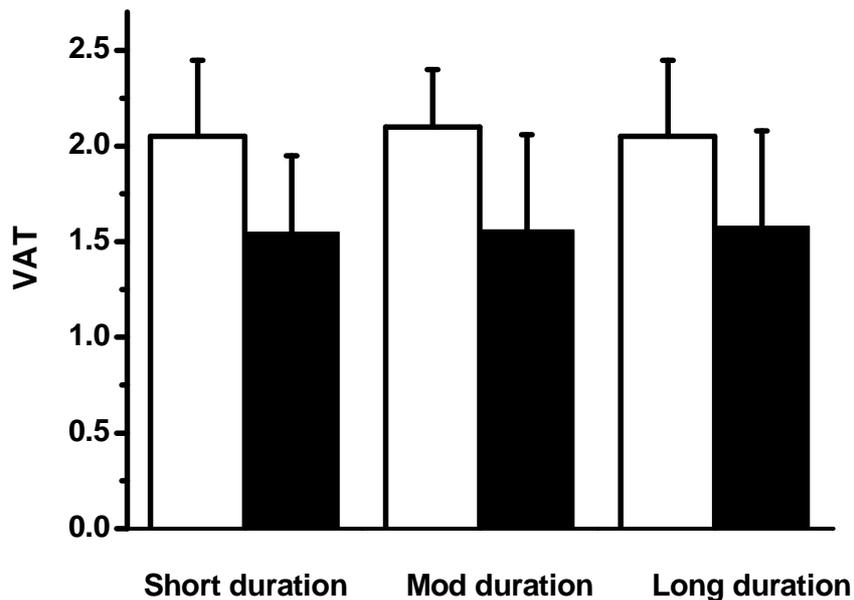


Figure 4-12 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data by $\dot{V}_E / \dot{V}_{O_2}$ method.

VAT $F_{ET}O_2\%$ from Edit data.

Figure 4.13 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 2.04 ± 0.40 (range 1.49-2.70) $l \cdot \text{min}^{-1}$ during the short protocol, 2.10 ± 0.33 (range 1.78-2.62) $l \cdot \text{min}^{-1}$ during the moderate protocol and 2.03 ± 0.40 (range 1.62-2.83) $l \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.55 ± 0.40 (range 1.16-2.41) $l \cdot \text{min}^{-1}$ during the short protocol, 1.55 ± 0.47 (range 1.01-2.36) $l \cdot \text{min}^{-1}$ during the moderate protocol and 1.58 ± 0.46 (range 1.04-2.29) $l \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

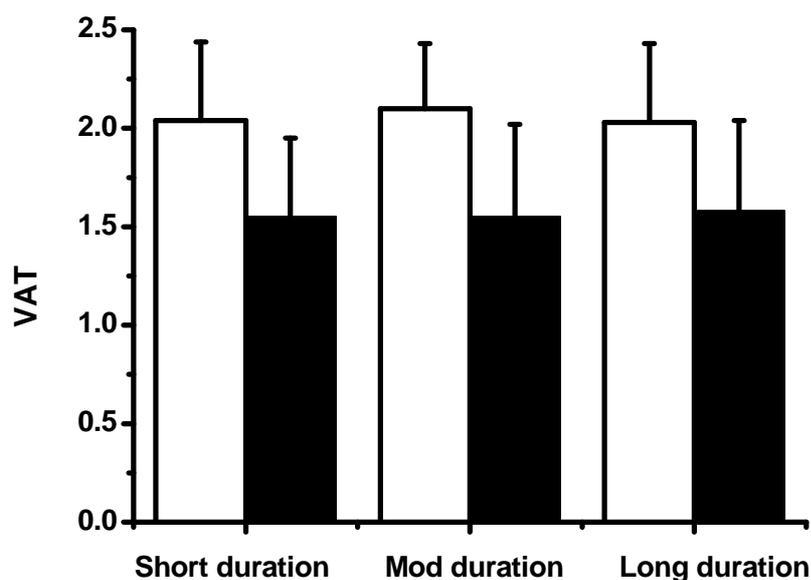


Figure 4-13 illustrates the impact of short, moderate and long test ramp protocols on the VAT during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data by $F_{ET}O_2\% / \dot{V}_{O_2}$ method.

Influence of test ramp protocols on treadmill work - with gas exchange data

Similarly, figure 4.14 shows that VAT was identified in male volunteers at a work load of 128 ± 18 (range 103-164) W during the short protocol, 115 ± 19 (range 73.70-145) W during the moderate protocol and 105 ± 16 (range 88.20-135) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 100 ± 18 (range 75.20-120) W during the short protocol, 90 ± 28 (range 59.70-124) W during the moderate protocol and 76 ± 19 (range 59.60-104) W during the long protocol. Like their male counterparts, there was no significant difference in the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

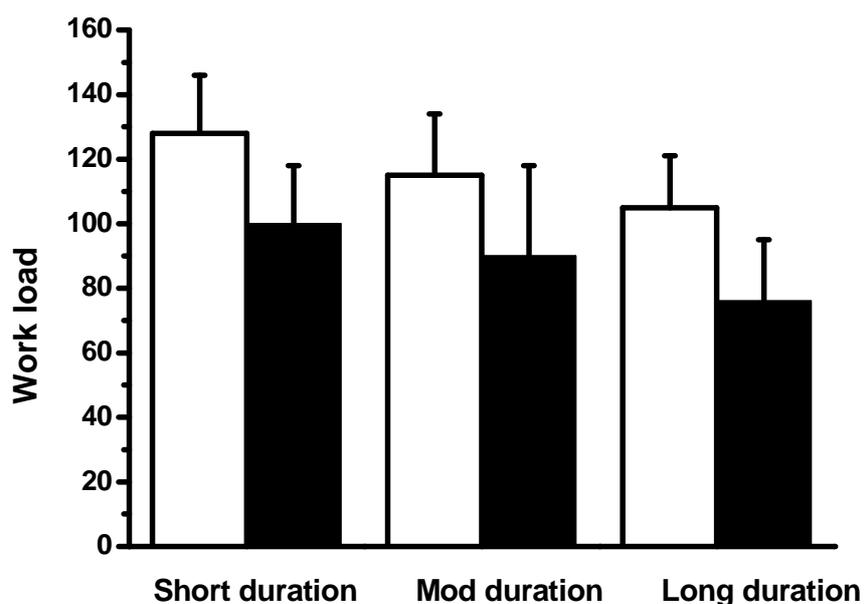


Figure 4-14 illustrates the impact of short, moderate and long test ramp protocols on the work load during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Influence of test ramp protocols on treadmill \dot{V}_E - with gas exchange data

Furthermore, figure 4.15 shows that VAT was identified in male volunteers at \dot{V}_E of 43.20 ± 9 (range 30.30-62) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 44.52 ± 7 (range 29-55.90) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 45.85 ± 6 (range 36-56) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at \dot{V}_E of 33.20 ± 5.60 (range 25-44) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 35.70 ± 10 (range 23-55) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 37.20 ± 9.50 (range 25-51) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

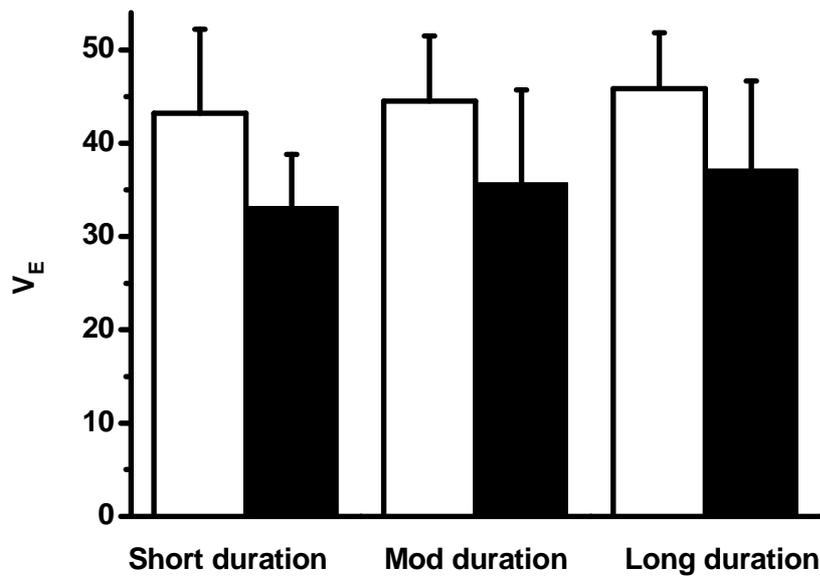


Figure 4-15 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during treadmill studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data by using respiratory compensation methods RCP

Influence of test ramp protocols on cycle ergometer VAT - with no data editing

Figure 4.16 illustrates the impact of short, moderate and long test ramp protocols (see figure 4.16 and associated text) on the VAT for male and female volunteers.

Figure 4.16 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.71 ± 0.30 (range 1.34-2.20) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.82 ± 0.30 (range 1.34-2.18) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.84 ± 0.20 (range 1.34-2.10) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.20 ± 0.20 (range 0.98-1.64) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.14 ± 0.20 (range 0.92-1.38) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.20 ± 0.30 (range 0.94-1.61) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

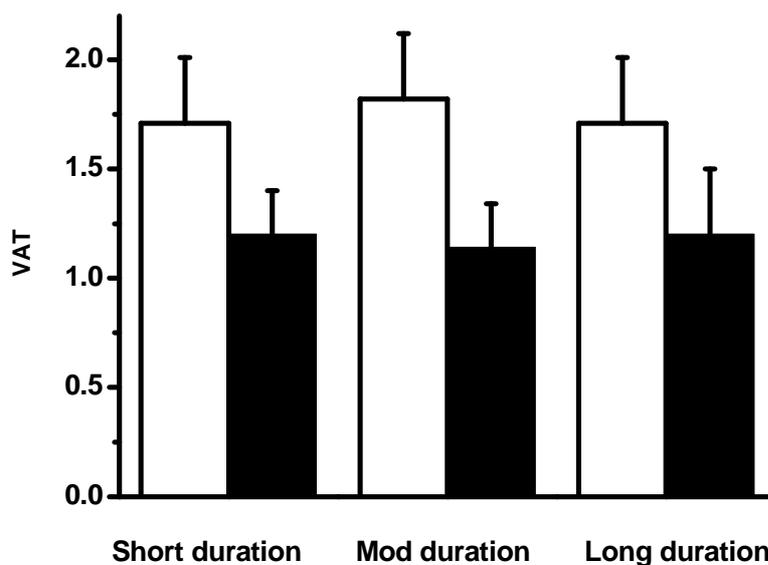


Figure 4-16 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Similarly, figure 4.17 shows that VAT was identified in male volunteers at a work load of 151 ± 51 (range 106-283) W during the short protocol, 117 ± 26 (range 723-161) W during the moderate protocol and 109 ± 19 (range 77-139) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 97 ± 37 (range 48-169) W during the short protocol, 80 ± 15 (range 65-106) W during the moderate protocol and 78 ± 11 (range 66-96) W during the long protocol. Like their male counterparts, there was no significant difference in the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

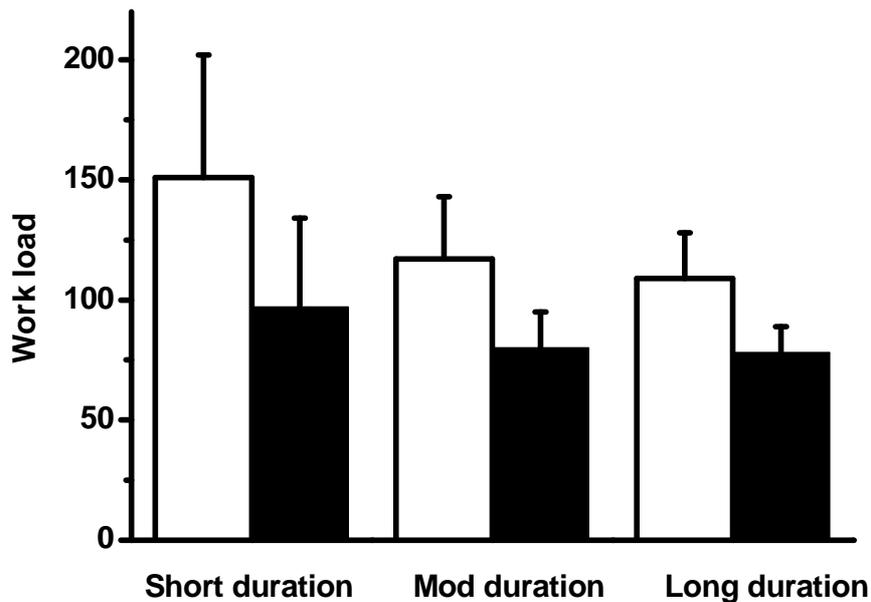


Figure 4-17 illustrates the impact of short, moderate and long test ramp protocols on the work load during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Furthermore, figure 4.18 shows that VAT was identified in male volunteers at \dot{V}_E of 37.90 ± 9 (range 23-50) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 41.1 ± 90 (range 28-51) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 45.4 ± 13 (range 22-64) $\text{l} \cdot \text{min}^{-1}$ during

the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at \dot{V}_E of 34.30 ± 10 (range 25-56) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 26.70 ± 3.60 (range 20-30) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 29.30 ± 6 (range 20-40) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

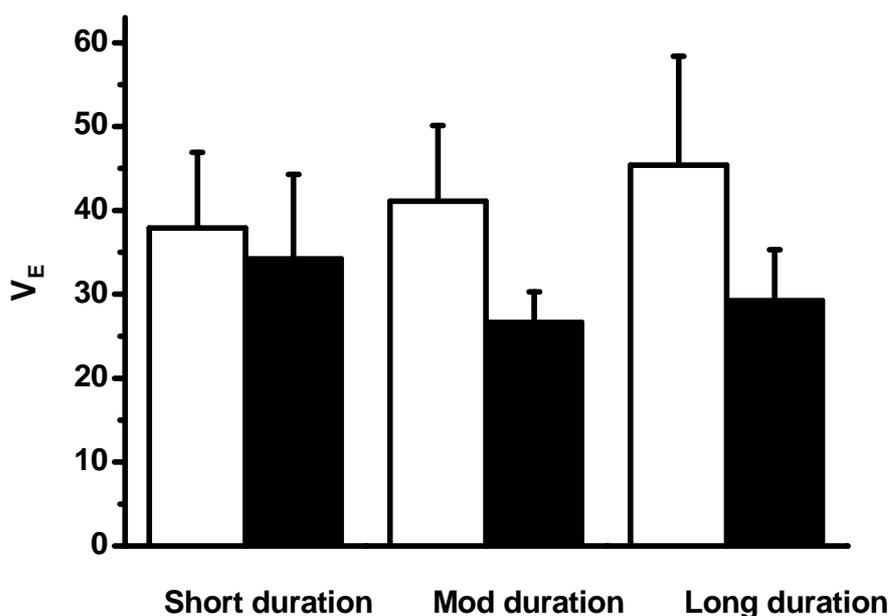


Figure 4-18 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Influence of test ramp protocols on treadmill VAT - with data editing

Figure 4.19 illustrates the impact of short, moderate and long test ramp protocols (see figure 2.3 and associated text) on the VAT for male and female volunteers.

Figure 4.19 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.71 ± 0.30 (range 1.27-2.15) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 1.78 ± 0.40 (range 1.31-2.22) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 1.79 ± 0.30 (range 1.33-2.28) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. There was no significant difference

in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.20 ± 0.20 (range 0.97-1.63) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.18 ± 0.22 (range 0.93-1.53) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.21 ± 0.19 (range 0.99-1.52) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

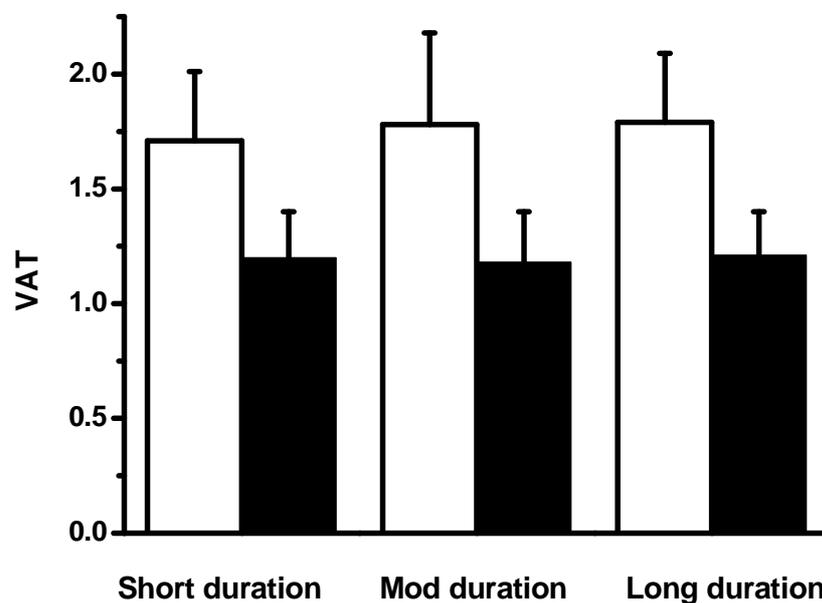


Figure 4-19 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Similarly, figure 4.20 shows that VAT was identified in male volunteers at a work load of 151 ± 51 (range 106-283) W during the short protocol, 117 ± 26 (range 723-161) W during the moderate protocol and 109 ± 19 (range 77-139) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 97 ± 37 (range 48-169) W during the short protocol, 80 ± 15 (range 65-106) W during the moderate protocol and 78 ± 11 (range 66-96) W during the long protocol. Like their male counterparts, there was no significant difference in

the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

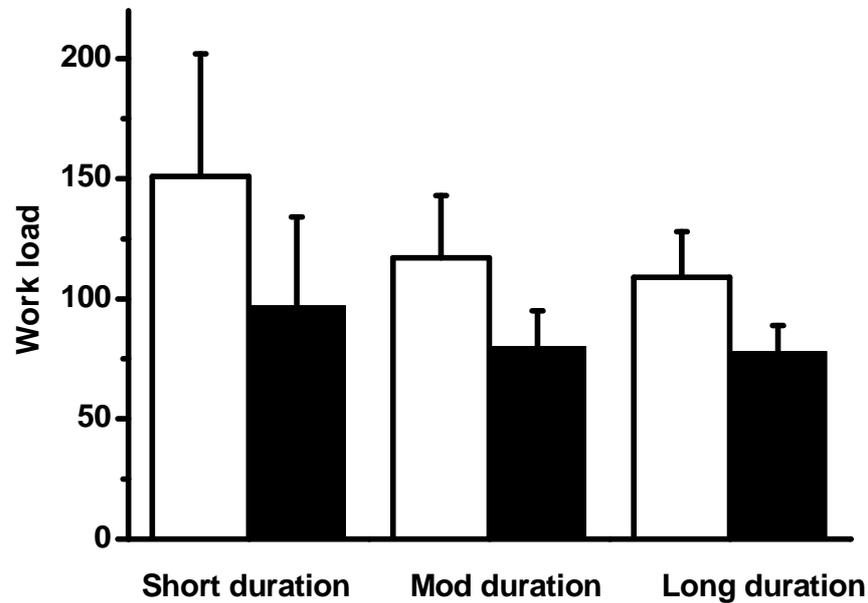


Figure 4-20 illustrates the impact of short, moderate and long test ramp protocols on the work load during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Furthermore, figure 4.21 shows that VAT was identified in male volunteers at \dot{V}_E of 38.70 ± 5 (range 29-46) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 40.49 ± 9 (range 29-56) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 43.23 ± 10 (range 23-58) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at \dot{V}_E of 34.20 ± 8 (range 27-52) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 26.43 ± 3 (range 21-30) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 29.31 ± 6 (range 20.30-41) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

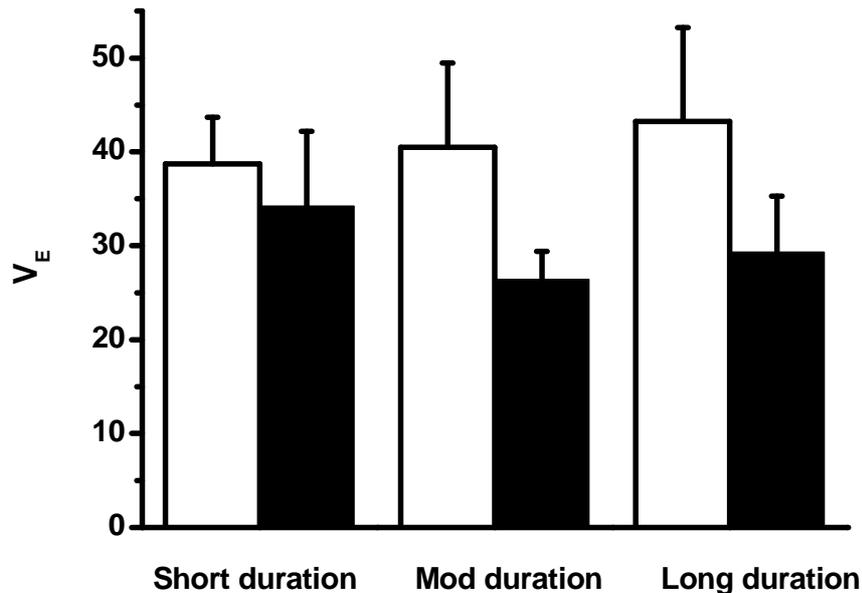


Figure 4-21 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data.

Influence of test ramp protocols on treadmill VAT - with gas exchange data

As previously stated, the use of gas exchange data allows the VAT to be identified without relying on the simple V-slope method and is therefore less sensitive to factors influencing linear regression analysis. For this reason, the influence of exercise duration can be tested on the \dot{V}_{O_2} , work load and \dot{V}_E at VAT directly rather than on artefacts of analysis.

Figure 4.22 illustrates the impact of short, moderate and long test ramp protocols (see figure 2.1 and associated text) on the VAT for male and female volunteers.

VAT by $\dot{V}_E / \dot{V}_{O_2}$ from Raw data.

Figure 4.22 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.91 ± 0.40 (range 1.35-2.60) l.min⁻¹ during the short protocol, 1.96 ± 0.50 (range 1.42-2.70) l.min⁻¹ during the moderate protocol and 1.87 ± 0.30 (range 1.35-2.50) l.min⁻¹ during the long protocol. There was no significant difference

in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.24 ± 0.23 (range 0.96-1.67) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 1.18 ± 0.17 (range 1.13-1.39) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 1.21 ± 0.21 (range 0.96-1.62) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

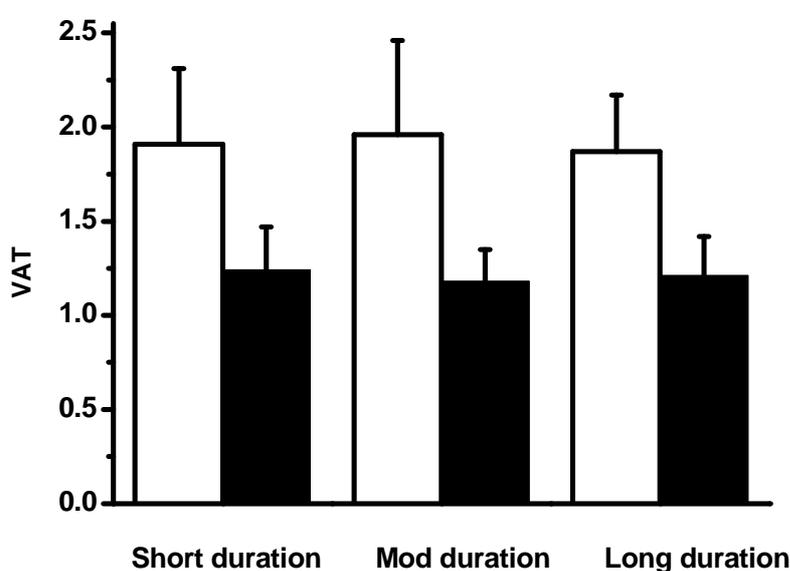


Figure 4-22 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data by $\dot{V}_E / \dot{V}_{O_2}$ method.

VAT $F_{ET}O_2\%$ from Raw data.

Figure 4.23 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.91 ± 0.41 (range 1.40-2.50) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 1.91 ± 0.50 (range 1.41-2.60) $\text{l}\cdot\text{min}^{-1}$ during the moderate protocol and 1.80 ± 0.29 (range 1.37-2.20) $\text{l}\cdot\text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.23 ± 0.24 (range 0.90-1.60) $\text{l}\cdot\text{min}^{-1}$ during the short protocol, 1.20 ± 0.17 (range 0.99-1.46) $\text{l}\cdot\text{min}^{-1}$

during the moderate protocol and 1.22 ± 0.21 (range 1.01-1.60) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

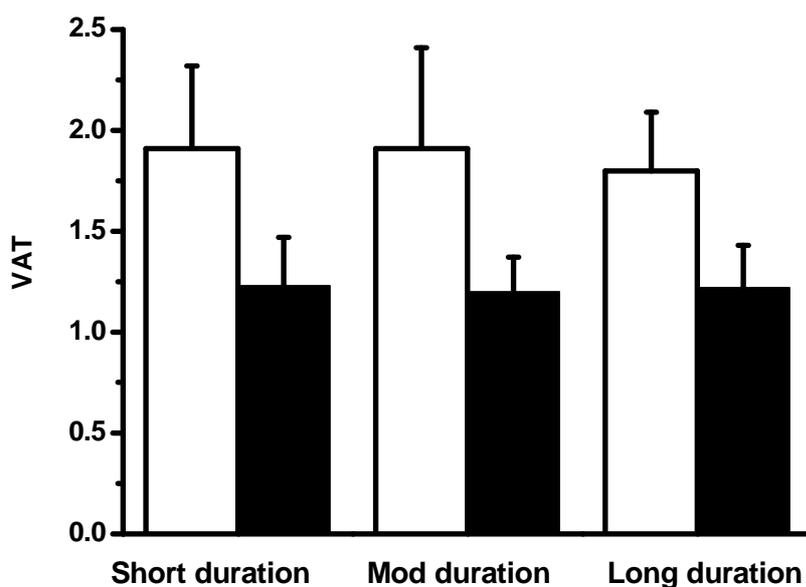


Figure 4-23 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data by $F_{\text{ET}}\text{O}_2\% / \dot{V}_{\text{O}_2}$ method.

VAT by $\dot{V}_E / \dot{V}_{\text{O}_2}$ from Edit data.

Figure 4.24 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.77 ± 0.34 (range 1.34-2.30) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.82 ± 0.35 (range 1.4-2.20) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.85 ± 0.33 (range 1.40-2.30) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.25 ± 0.25 (range 1.00-1.70) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 1.18 ± 0.22 (range 0.93-1.53) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 1.27 ± 0.20 (range 1.01-1.60) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in

the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols. However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

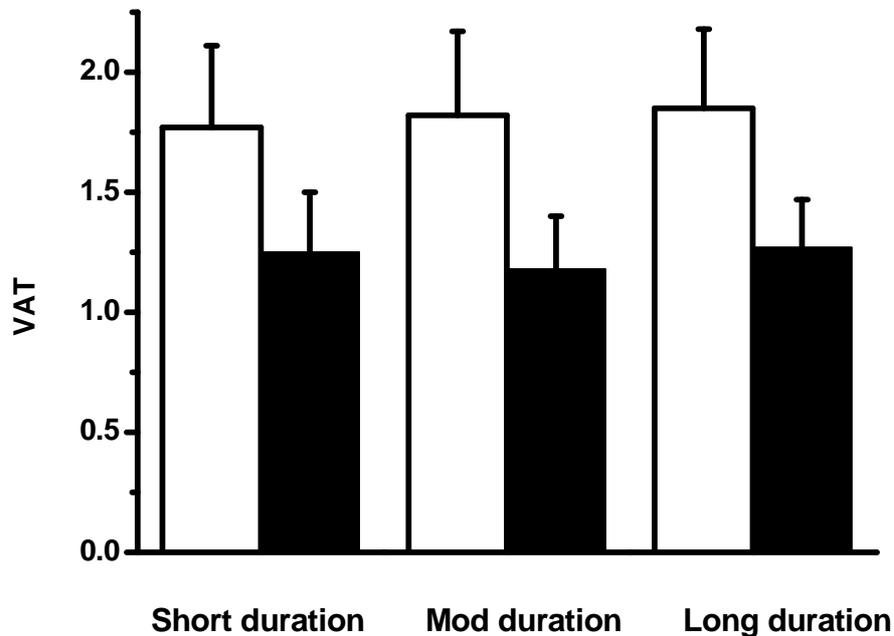


Figure 4-24 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data by $\dot{V}_E / \dot{V}_{O_2}$ method.

VAT $F_{ET}O_2\%$ from Edit data.

Figure 4.25 shows that VAT was identified in male volunteers at an absolute \dot{V}_{O_2} of 1.75 ± 0.34 (range 1.30-2.20) $l \cdot \text{min}^{-1}$ during the short protocol, 1.82 ± 0.36 (range 1.40-2.30) $l \cdot \text{min}^{-1}$ during the moderate protocol and 1.85 ± 0.32 (range 1.40-2.30) $l \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at an absolute \dot{V}_{O_2} of 1.24 ± 0.24 (range 0.99-1.70) $l \cdot \text{min}^{-1}$ during the short protocol, 1.22 ± 0.21 (range 0.94-1.60) $l \cdot \text{min}^{-1}$ during the moderate protocol and 1.26 ± 0.21 (range 1.00-1.60) $l \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in the absolute \dot{V}_{O_2} measured at VAT in the females for any test ramp protocols.

However, the female absolute \dot{V}_{O_2} at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

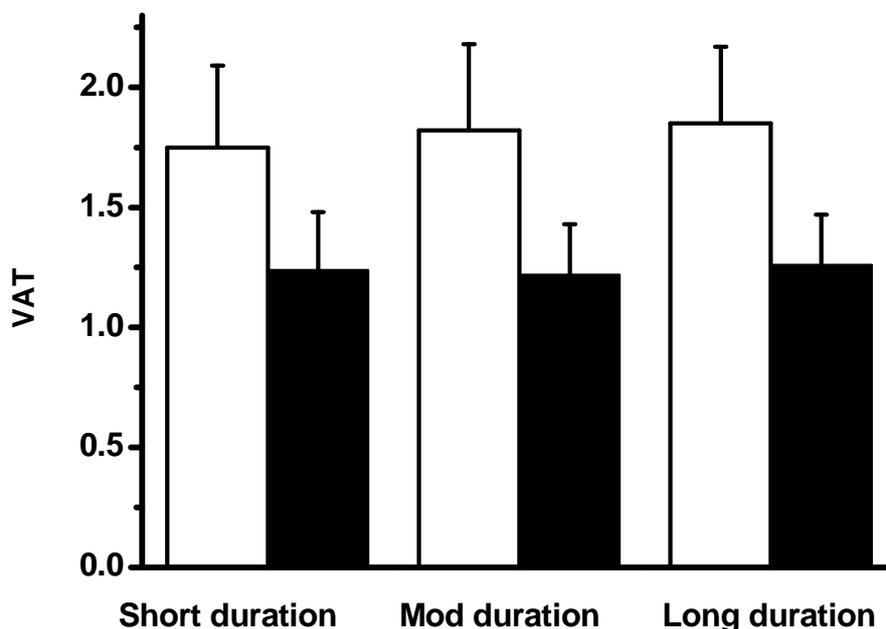


Figure 4-25 illustrates the impact of short, moderate and long test ramp protocols on the VAT during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from edited data by $F_{ET}O_2\% / \dot{V}_{O_2}$ method.

Similarly, figure 4.26 shows that VAT was identified in male volunteers at a work load of 151 ± 51 (range 106-283) W during the short protocol, 117 ± 26 (range 723-161) W during the moderate protocol and 109 ± 19 (range 77-139) W during the long protocol. There was no significant difference in the work load at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at a work load of 97 ± 37 (range 48-169) W during the short protocol, 80 ± 15 (range 65-106) W during the moderate protocol and 78 ± 11 (range 66-96) W during the long protocol. Like their male counterparts, there was no significant difference in the work load at VAT in the females for any test ramp protocols. However, the female work load at VAT was significantly lower than the male during the test ramp protocols ($P < 0.05$, repeated measures).

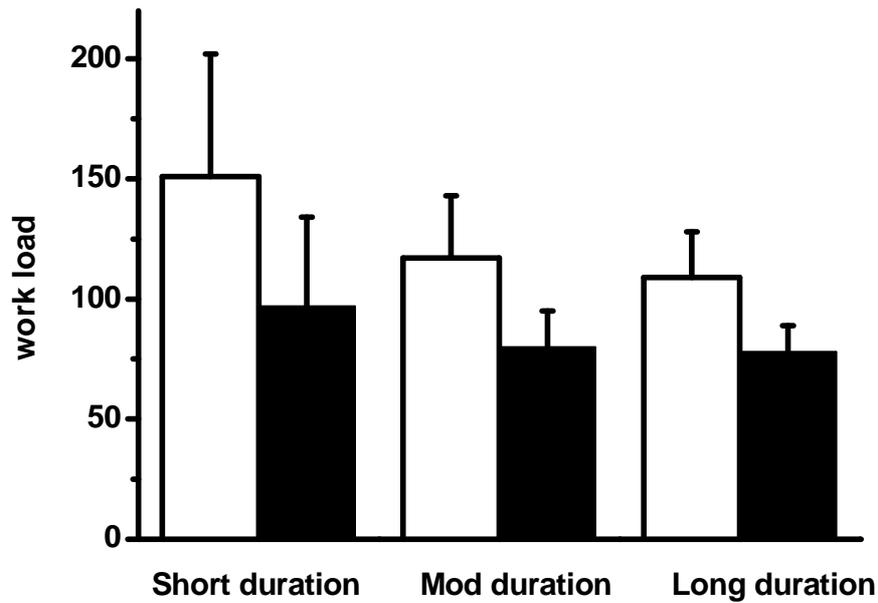


Figure 4-26 illustrates the impact of short, moderate and long test ramp protocols on the work load during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars which is measured from raw data.

Furthermore, figure 4.27 shows that VAT was identified in male volunteers at \dot{V}_E of 39.47 ± 6.93 (range 28-52) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 41.94 ± 8.62 (range 31-56) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 43.53 ± 9.24 (range 28-57.8) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. There was no significant difference in \dot{V}_E at VAT in the males for any test ramp protocols. VAT was identified in female volunteers at \dot{V}_E of 32.31 ± 5.26 (range 25.70-42) $\text{l} \cdot \text{min}^{-1}$ during the short protocol, 33.32 ± 4.85 (range 29-44.5) $\text{l} \cdot \text{min}^{-1}$ during the moderate protocol and 33.24 ± 7.44 (range 25.50-50) $\text{l} \cdot \text{min}^{-1}$ during the long protocol. Like their male counterparts, there was no significant difference in \dot{V}_E at VAT in the females for any test ramp protocols.

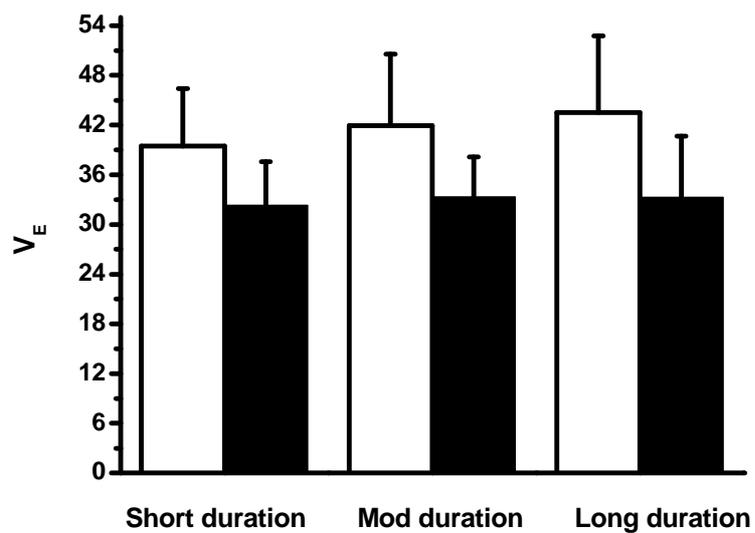


Figure 4-27 illustrates the impact of short, moderate and long test ramp protocols on the \dot{V}_E during cycle ergometer studies, Male volunteers are represented by the open bars and female volunteers by the closed bars from RCP data.

4.7 Discussion

This study was undertaken to examine the impact of exercise duration on the determination of the ventilatory threshold. This aim was achieved by re-programming the duration of individual work rate increments in a ramp Bruce protocol (Will & Walter, 1999). This approach meant that the speed/gradient increments in the treadmill tests and the resistance increments on cycle ergometer tests were constant and the only changing variable was exercise duration. Consequently, there was no significant difference in the absolute oxygen uptake, work load or ventilation at the ventilatory threshold in either exercise modality studied. This is an important finding since the determination of a range of sub-maximal measures of exercise performance are influenced by the position of the ventilatory threshold.

The ventilatory threshold is a useful non-invasive measure that is related to the lactate threshold (Sinclair *et al.*, 2009). The lactate threshold is the point where lactate removal in the working muscle is unable to keep up with the lactate production. During the demands of high-intensity exercise, the muscle uses glucose and glycogen as metabolic substrates for aerobic energy metabolism (glycolysis). The final step of glycolysis results in the production of two molecules of pyruvate which accumulates in the cell and absorbs two protons released from ATP hydrolysis to form lactate. Thus, lactate production is a consequence of muscle acidosis rather than a cause and acts as a temporary 'buffer' of the accumulated protons. Since increased lactate production coincides with the acidosis, lactate measurement is an excellent 'indirect' marker for the metabolic condition of the muscle. Thus the increase in blood lactate is associated with a metabolic acidosis that is also 'buffered' by bicarbonate in the blood stream which releases carbon dioxide and stimulates ventilation to allow respiratory compensation to help maintain homeostatic control of blood pH. Thus the ventilatory threshold can be observed as an increase in carbon dioxide production relative to oxygen uptake in expired gas analysis which follows shortly after the lactate threshold (there is a short time delay for chemoreceptor activation) (Wasserman *et al.*, 1985; Wasserman *et al.*, 1973).

This means that a simple plot of the relationship between carbon dioxide production (\dot{V}_{CO_2}) and oxygen uptake (\dot{V}_{O_2}), the so-called V-slope, can provide useful information on the relative intensity of exercise being undertaken and provide a measure of metabolic stress/efficiency (Caiozzo *et al.*, 1982). The ventilatory threshold can be identified as a breakpoint in the linear $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship at the intercept of the S_1 and S_2 components (see figure 4.28 and associated text). However, this simple relationship can be influenced by a number of experimental artefacts that make its accurate identification difficult (Hopker *et al.*, 2011).

For example, volitional or anticipatory hyperventilation before exercise will alter the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship through non-metabolic production of CO_2 and can lead to the phenomenon known as a pseudo-threshold (Whipp, 2007). This artefact can be reduced by not starting cardiopulmonary exercise testing if the patient's resting RER is in excess of 0.95 and introducing a period of unloaded work at the beginning of the protocol (as was done in this study). However, even with these additional procedures it is possible that the early stages of exercise (the cardio-dynamic phase) may be associated with non-metabolic production of CO_2 that can influence the accuracy of the breakpoint determination.

In addition, visual identification of VAT by the V-Slope method was an intricate procedure, specially for data derived from long exercise duration protocols. Figure 4.28 illustrates an example of this problem. The data comes from a very fit 42 year male subject (LJ) who is a regular exerciser (>6 years endurance activity). In this subject, the break point in the V-slope could not be seen in longer duration exercise (figure 4.28 A). However, at moderate (figure 4.28, B) and short (figure 4.28, C) duration exercise the break point is seen more clearly.

Despite the limitations in the V-slope, it is possible to establish the ventilatory threshold, even during long duration exercise, using standard gas exchange criteria to facilitate the visual interpretation of the break point (figure 4.29).

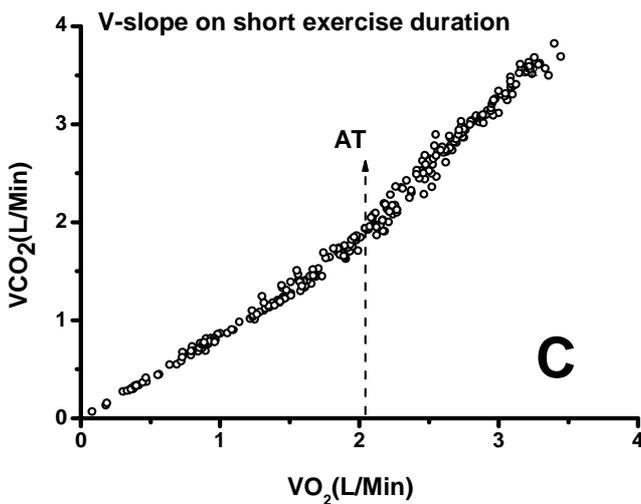
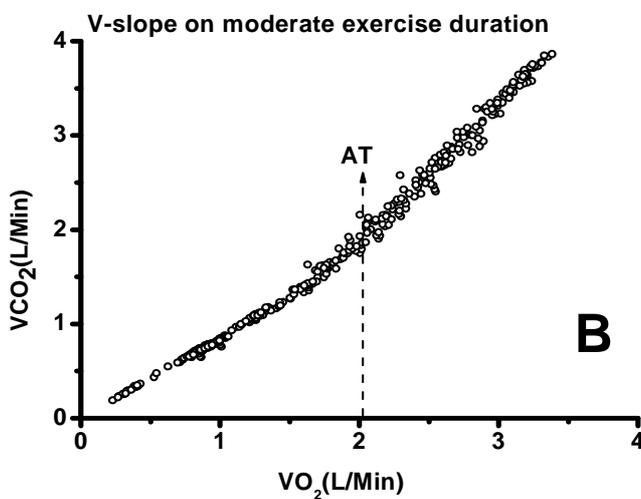
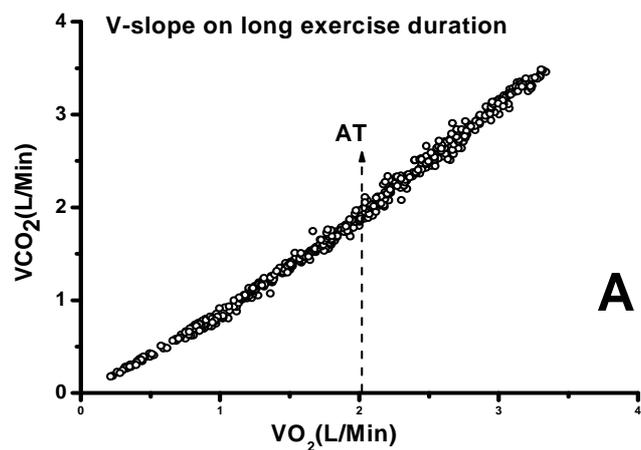


Figure 4-28 Effects of exercise duration on visual determination of the V-slope break point in a single subject.

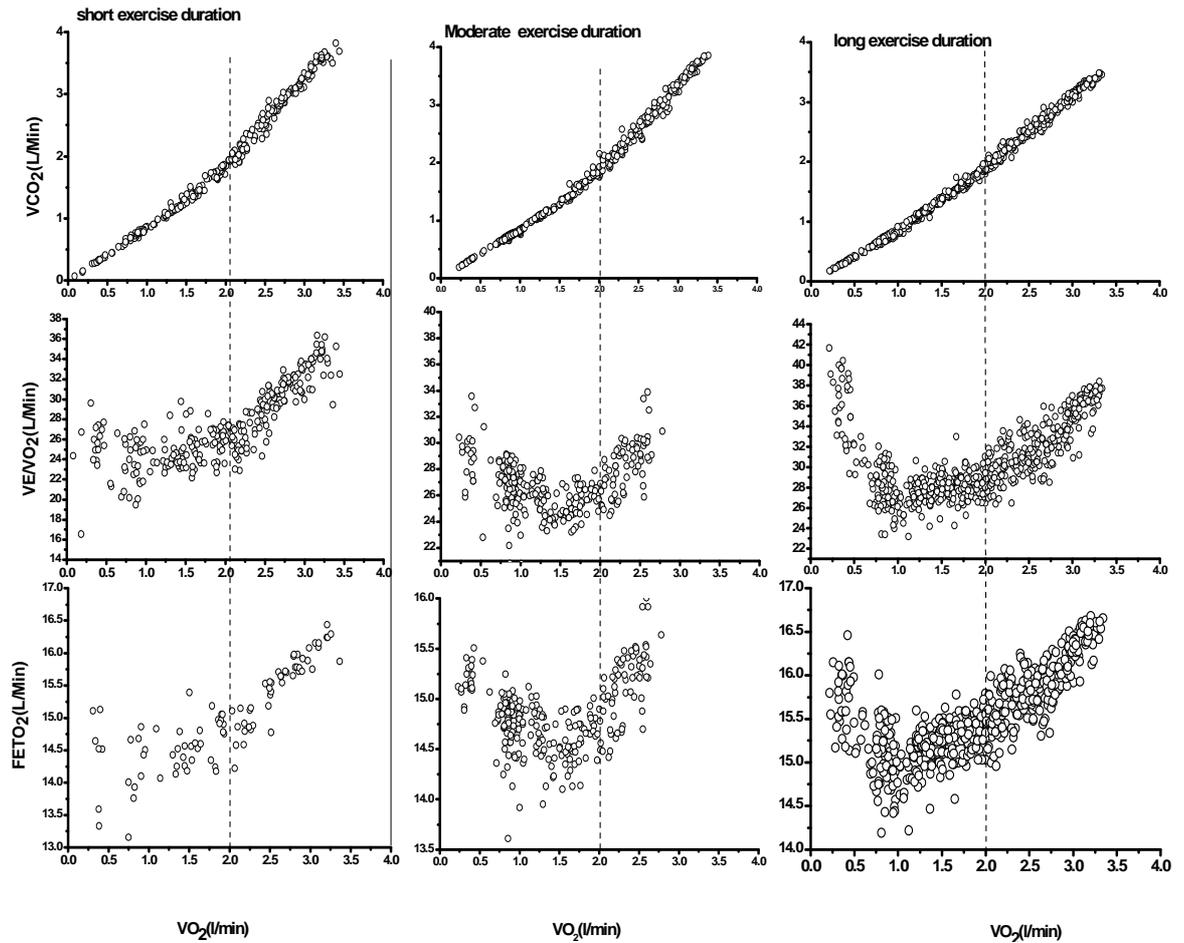


Figure 4-29 Determining the ventilation at anaerobic threshold using standard gas exchange criteria.

The rationale behind this current study was that changing the duration of exercise could influence the slope of the S_1 component and subsequently compromise the validity of the breakpoint determination in the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship. Figure 4.30 shows that during short duration exercise, a greater proportion of the data obtained to allow a fit of the S_1 component would be associated with the cardio-dynamic phase and if there is non-metabolic production of CO_2 then the higher levels would reduce the slope of the S_1 component and result in a lower oxygen uptake at the point of intercept with the S_2 component. Similarly, with long duration the change in the relative proportion of data contributing to the S_1 and S_2 components may make it difficult to visualise a breakpoint in the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship (shown as a higher oxygen uptake at the point of intercept in this illustration).

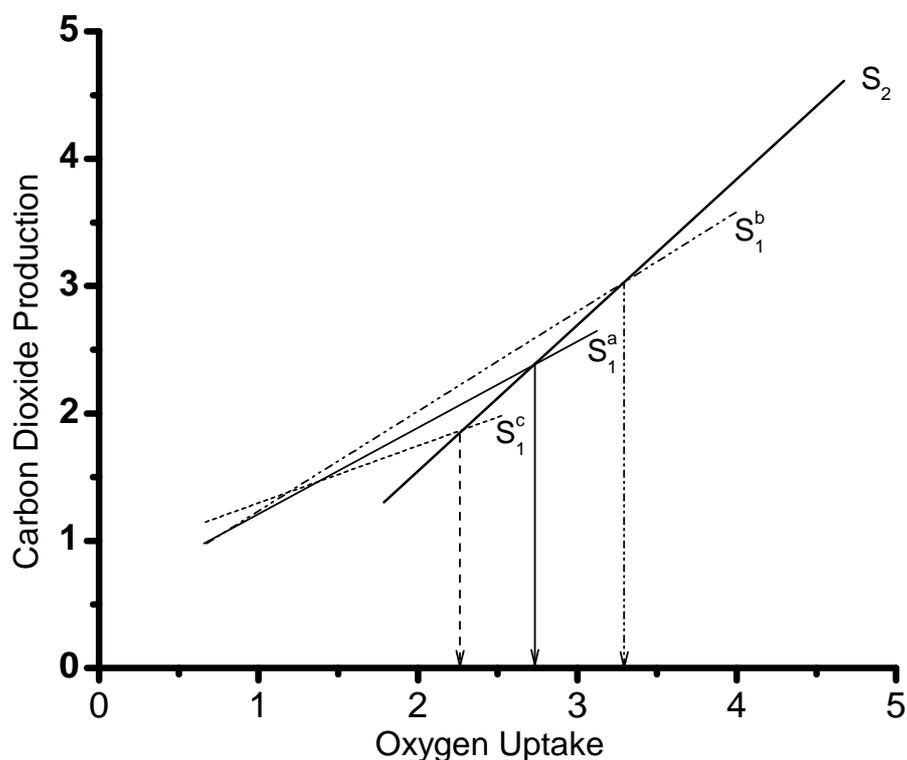


Figure 4-30 An illustration showing the potential effects of exercise duration on the S_1 and S_2 components of the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship. The superscript characters a (with a solid line), b (with a dashed and dotted line) and c (with a dashed line) show the effect of changing the slope of the S_1 component on the position of the breakpoint in the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship. The character a represents moderate duration exercise, b long duration exercise and c short duration exercise.

This study bears out this rationale in that the slope and fit (the R^2 and RSS measurements) of the S_1 component was reduced during short duration exercise. However, the functional relevance of this statistical analysis of the S_1 component is limited because the reduced slope did not result in statistically significant changes in the physiologically relevant parameters measured at the breakpoint determined by the simple V-slope method. However, while both the slope and the fit of the S_1 component increased with exercise duration it was difficult, in some cases, to visualise a clear breakpoint in the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship during long duration exercise (see figure 4.28). This could limit the validity of the simple V-slope measurement in some patients when using longer duration protocols, but it is generally valid since there were no statistically

significant changes in the physiologically relevant parameters measured at the breakpoint determined by the simple V-slope method.

Thus, most automated gas analysis systems that are used in clinical practice will provide an accurate assessment of the ventilatory threshold when exercising for shorter or longer than the current ACSM guidelines for clinical exercise testing. No specific guidelines are required to determine the ventilatory threshold when following a protocol that aims to elicit a maximal response within eight to twelve minutes. While in longer duration exercise there may be some patients that provide challenges in identifying a valid breakpoint in the $\dot{V}_{CO_2}/\dot{V}_{O_2}$ relationship it is possible to accurately identify the ventilatory threshold by using truncated and edited data with additional gas exchange criteria. This level of analysis may not be easy to perform on some automated metabolic carts and in such circumstances it may be prudent to perform a shorter duration test. In the heterogeneous group (in terms of their age, gender, race and fitness levels) studied here it appears that there is minimal benefit from data editing procedures in determining a valid ventilatory threshold.

Wasserman and McIlroy were the first scientists to use the ventilatory anaerobic threshold to assess the functional capacity of cardiac patients (Wasserman & McIlroy, 1964). Clinicians often prefer to use sub-maximal markers of functional capacity due to the compliance and safety issue (Beaver *et al.*, 1986). However, sometimes it can be difficult to establish break point in the breath by breath data from frail patients (Hopker *et al.*, 2011). Despite this, sub-maximal exercise testing has been used in heart failure (NYHA Class II-III) to determine functional capacity (Lipkin *et al.*, 1985) and has been used for assessing pre-operative cardio-respiratory fitness in patients with abdominal aortic aneurysms (Kothmann *et al.*, 2009). In addition, anaerobic threshold $<11\text{ml.kg.min}^{-1}$ during (CPET) is used as a test to predict pre-operative risk in elderly patients (Sinclair *et al.*, 2009). It is now generally accepted that this measurement provides useful clinical information.

The outcomes from this chapter will now allow us to determine the effects of exercise duration *per se* on other clinically relevant markers of sub-maximal exercise performance in the following chapters.

CHAPTER FIVE

Oxygen Uptake Efficiency Slope

5 Impact of Exercise Duration on Oxygen Uptake Efficiency Slope

5.1 Introduction

The relationship between ventilation (\dot{V}_E) and oxygen uptake (\dot{V}_{O_2}) is linear before the anaerobic threshold but after this point the increase in \dot{V}_E is disproportionate to \dot{V}_{O_2} (Astrand & Rodahl, 1986; Wasserman K, 1999). The ratio of $\dot{V}_E / \dot{V}_{O_2}$ is representative of ventilatory efficiency and is normally around 25- 30 at rest (ATS/ACCP, 2003). However, this ratio is not linear with exercise intensity and falls during sub-maximal effort at low exercise intensities before rising to a maximum of around 35 at the end of a maximal effort in a healthy male (ATS/ACCP, 2003). The non linearity of this relationship has been addressed by Baba *et al* (1996) (Baba *et al.*, 1996) and they suggested the use of a semi-log transformation to derive a linear relationship that has been termed the oxygen uptake efficiency slope (OUES) (figure 5.1). This transformation is becoming a more frequently used sub-maximal marker of exercise performance.

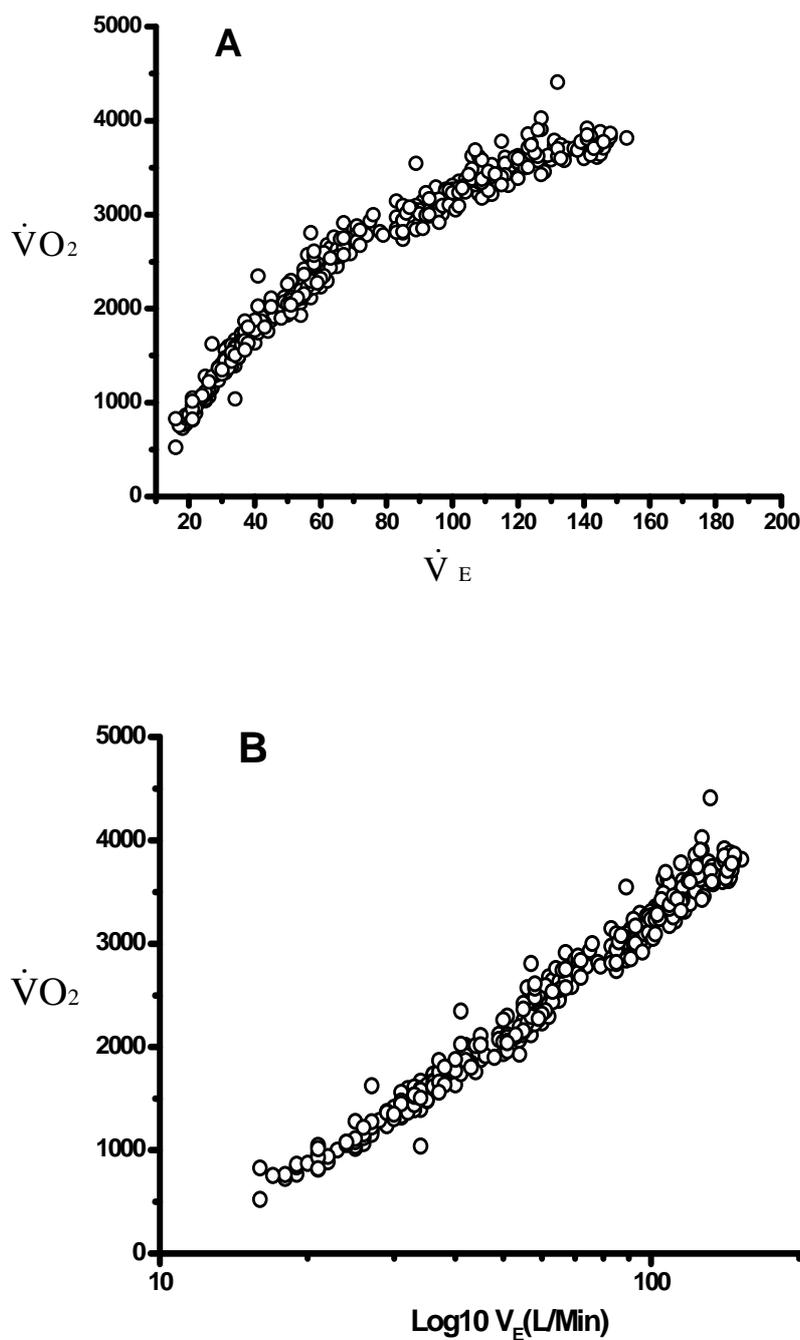


Figure 5-1 These plots illustrate the benefits of using a semi-log plot to provide a linear relationship between ventilation and oxygen uptake. The slope before (A) and after (B) log plot of \dot{V}_E L·min⁻¹ against $\dot{V}O_2$ ml⁻¹·kg⁻¹·min⁻¹.

The concept behind the OUES is quite simple; it measures the efficiency of the cardiopulmonary system to deliver oxygen to working muscle and, as such, a greater slope indicates greater ventilatory efficiency.

However, at the beginning of an activity effort, the oxygen consumption does not increase in proportion to work rate because of the use of immediate energy stores to support anaerobic metabolism. This is often referred to as the cardio-dynamic phase and can be a confounding variable in breath by breath gas analysis. For this reason, it has been recommended that the first 2-3 minutes of breath by breath data collection are excluded from data analysis to avoid the influence of the cardio-dynamic phase (Balady *et al.*, 2010). However, such manoeuvres are not incorporated into the measurement of OUES and during short duration exercise tests the cardio-dynamic phase may become a significant proportion of the total exercise duration.

The validity of OUES as an objective marker of functional capacity has been tested in many studies and it has been observed that OUES correlates strongly with $\dot{V}_{O_2 \text{ MAX}}$ and is not influenced by exercise intensity. In 2000, Hollenberg and Tager extended the initial work of Baba *et al* (1996) to show that OUES is a valid and reliable index of cardio-respiratory functional capacity in older adults (Hollenberg & Tager, 2000). This study employed 998 older adults without clinical evidence of cardiovascular disease and a smaller sample of patients with congestive heart failure (CHF), n=12. Their findings indicate that when OUES is calculated from the first 75% of total exercise duration then the value was not significantly different from that observed over full exercise duration. The implication being that sub-maximal testing and the concomitant risk reduction was a useful surrogate for the maximum oxygen consumption.

In 1999, Baba *et al* also employed the OUES in the evaluation of adult cardiac patients with chronic heart failure (Baba *et al.*, 1999). Their findings similarly showed that the OUES obtained through different exercise intensities was consistent and correlated well with $\dot{V}_{O_2 \text{ MAX}}$. Additionally, OUES was found to be an effective discriminator of New York Heart Association functional class. These findings suggest that OUES could be a prognostic indicator in addition to being a valid measure of functional capacity.

Pichon *et al* (2002) reported that there were inter-individual variations between $\dot{V}_{O_2 \text{ MAX}}$ and OUES despite a significant correlation and have suggested that OUES might be limited in clinical practice (Pichon *et al.*, 2002). However, these

authors acknowledged that OUES is a valid sub-maximal index of cardio-respiratory reserve that can classify individuals according to their functional capacity. They emphasised that the purpose of their study was to evaluate the inter-changeability of OUES and $\dot{V}_{O_2 \text{ MAX}}$ rather than an assessment of its use in functional assessment. The OUES has been found to be a useful indicators of functional capacity in frail or young patients (Baba *et al.*, 1996; Berger *et al.*, 2011; Pinkstaff *et al.*, 2010), in the obese (Drinkard *et al.*, 2007) and is reliable and reproducible in healthy adults (Van Laethem *et al.*, 2009).

Combined, these data suggest that the OUES has specificity to predict functional capacity but is not an accurate/sensitive predictor of aerobic capacity. It is possible that this lack of sensitivity is due in part to the inclusion of the cardio-dynamic phase in the measurement of OUES. Moreover, the many studies that have calculated OUES tend to restrict the data analysis to around 70% of total exercise duration (Hollenberg & Tager, 2000) (Van Laethem *et al.*, 2005) (Mollard *et al.*, 2008). This will mean that much of the data published on OUES will include a relatively high proportion of the cardio-dynamic phase in the analyzed data and that the impact of the lactate threshold on the ventilatory response may be significantly reduced in some individuals (so that both factors may contribute to reduced sensitivity of the OUES to predict aerobic capacity). In other words, at 75% of total exercise duration the amount of data generated above the lactate threshold may be quite small.

Therefore, the aims of the current study are to determine the effect of exercise duration on OUES in the presence and absence of the cardio-dynamic phase and to establish the influence of restricted data analysis on the measured OUES. The data being analysed on the following basis:

- for the total exercise duration (OUES_{total})
- for 75% of the total exercise duration (OUES_{75%})
- for data before the lactate threshold (OUES_{before})
- for data after the lactate threshold (OUES_{after}).

5.2 Methods

Subjects

The main aim of this study is to address the issue of exercise duration and analysis has been restricted to volunteers that managed to complete three test ramp protocols on either the treadmill or cycle ergometer. Some of the volunteers managed to complete the necessary test ramp protocols on both modalities while others only managed this for one. The physical characteristics for the two groups are shown in table 5.1.

Variable	Gender	Treadmill	Cycle Ergometer
Number of Participants	Male	11	11
	Female	9	9
Age (years)	Male	31.2±6.0 (24-42)	32.5±6.0 (25-42)
	Female	36.6±11.0 * (24-57)	35.4±10.0 * (24-57)
Weight (kg)	Male	76.0±7.7 (60-84)	79.0±12.5 (60-103)
	Female	60.7±9.0 * (48-77)	60.0±9.0 * (48-77)
Height (cm)	Male	175.0±6.4 (163-182)	175.4±5.7 (163-182)
	Female	162.0±9.0 * (146-175)	160.7±7.3 * (145-172)
BSA [†] (m ²)	Male	1.92±0.14 (1.94-2.02)	1.96±0.12 (1.65-2.22)
	Female	1.65±0.14 * (1.47-1.87)	1.64±0.13 * (1.47-1.87)
BMI [#] (kg.m ⁻²)	Male	24.8±2.0 (21-28)	26.0±4.0 (21-34)
	Female	23.1±3.8 * (18-29)	23.0±4.0 * (18-29)

Table 5.1 Physical characteristics of all study volunteers. Data are presented as mean±SD (where appropriate) and the range is given in parenthesis. * indicates $P < 0.05$ compared with males (t -test with Bonferroni correction). [†] BSA is Body Surface Area according to Mosteller formula. [#] BMI is Body Mass Index.

5.3 Calculation of Oxygen Uptake Efficiency Slope

The \dot{V}_{O_2}/\dot{V}_E relationship was described by the following equation to obtain a value for OUES;

$$\dot{V}_{O_2} = a \text{ Log}_{10} \dot{V}_E + b \quad \text{equation (5.1)}$$

Where a represents the constant describing the rate of \dot{V}_E increase necessary to maintain the required \dot{V}_{O_2} and provides a value for the OUES (Baba *et al.*, 1996).

5.4 Statistical analysis

All data are presented as mean \pm standard deviation (SD) and all statistical analyses were performed using Origin Pro 8 SR4 v8. 0951(B951) and SPSS (18.0.0. Chicago) for each case and linear regression were derived to determine the OUES. At the 0.05 level, all data are normally distributed by Shapiro-Wilks normality test and suitable for parametric data analysis. All OUES_{total}, OUES_{before}, OUES_{after} and OUES_{75%} in both exercise modalities were significantly drawn from a normal distributed population. A Repeated measurement ANOVA was used to examine the differences between OUES_{total}, OUES_{before}, OUES_{after} and OUES_{75%}. If a significant difference was predicted then a *post hoc* test with Bonferroni adjustments was used for multiple comparisons. Statistical significance was analysed using appropriate *t*-tests and set at the 5% level. Pearson's correlation coefficients were employed to measure the linear relation (with 95% CI) between OUES and \dot{V}_{O_2} .

5.5 Results

Physical characteristics

Table 5.1 indicates that the study participants are heterogeneous showing a wide range in age and body habitus. However, there was no significant difference in the physical characteristics between the two exercise modality groups. As expected, the male volunteers were significantly taller and heavier (resulting in higher BSA and BMI) than the female volunteers. The female group were significantly older than the males.

5.6 Influence of test ramp protocols on exercise duration

The different biomechanics and energy demands of walking and cycling can influence cardiopulmonary measurements during maximal effort. Consequently, the data for each exercise modality are presented separately.

Influence of test ramp protocols on treadmill $OUES_{total}$ (including cardio-dynamic phase).

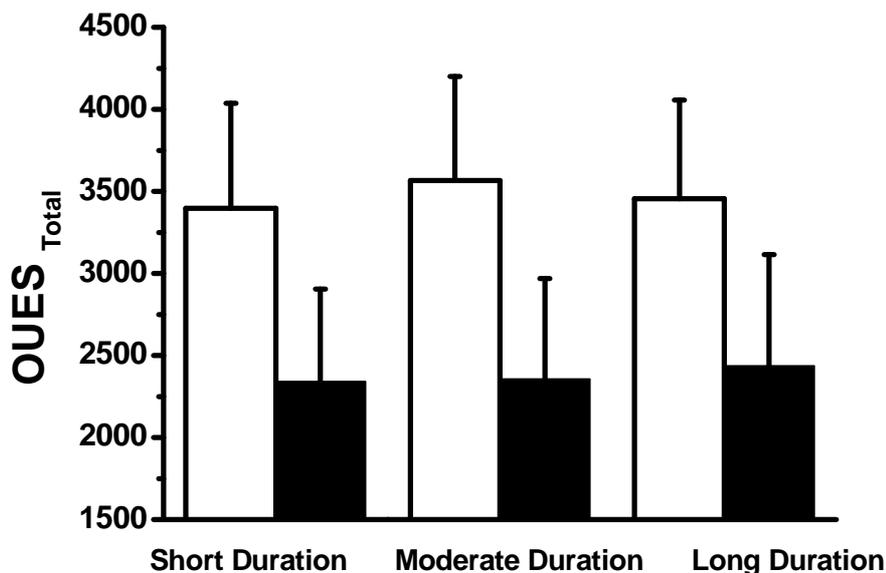


Figure 5-2 illustrates total oxygen uptake efficiency slope $OUES_{total}$ including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.2 shows the impact of short, moderate and long test ramp protocols on the $OUES_{total}$ determined in the male and female volunteers. Males achieved $OUES_{total}$ of 3398 ± 639 (range 2263-4332) during the short protocol, 3565 ± 635 (range 2411-4757) during the moderate protocol and 3456 ± 600 (range 2282-4471) during the long protocol. There was no significant difference in the $OUES_{total}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{total}$ of 2341 ± 564 (range 1611-3003) during the short protocol, 2356 ± 613 (range 1567-3149) during the moderate protocol and 2436 ± 679 (range 1606-3843) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{total}$ achieved by the females in the test ramp protocols. However, the female $OUES_{total}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{total}$ (excluding cardio-dynamic phase).

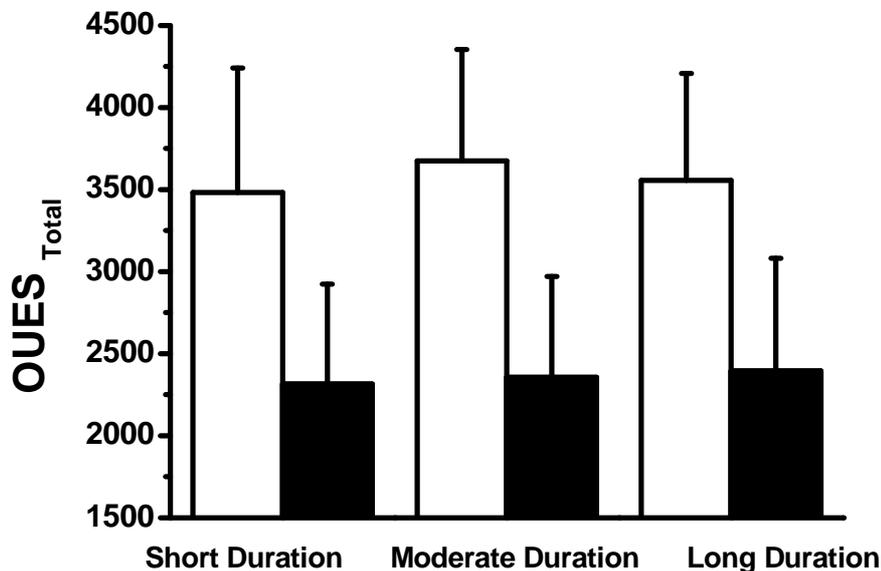


Figure 5-3 illustrates total oxygen uptake efficiency slope $OUES_{Total}$ excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.3 shows the impact of short, moderate and long test ramp protocols on the $OUES_{total}$ determined in the male and female volunteers. Males achieved

OUES_{total} of 3482±760 (range 2197-4413) during the short protocol, 3675±680 (range 2453-4981) during the moderate protocol and 3557±650 (range 2263-4659) during the long protocol. There was no significant difference in the OUES_{total} achieved by the males in the test ramp protocols. Females achieved OUES_{total} of 2316± 607 (range 1598-3422) during the short protocol, 3677±680 (range 2453-4981) during the moderate protocol and 2396±686 (1626-3420) during the long protocol. Like their male counterparts, there was no significant difference in the OUES_{total} achieved by the females in the test ramp protocols. However, the female OUES_{total} was significantly lower than the male during all test ramp protocols ($P<0.05$, t -test).

Influence of test ramp protocols on treadmill OUES_{75%} (including cardio-dynamic phase).

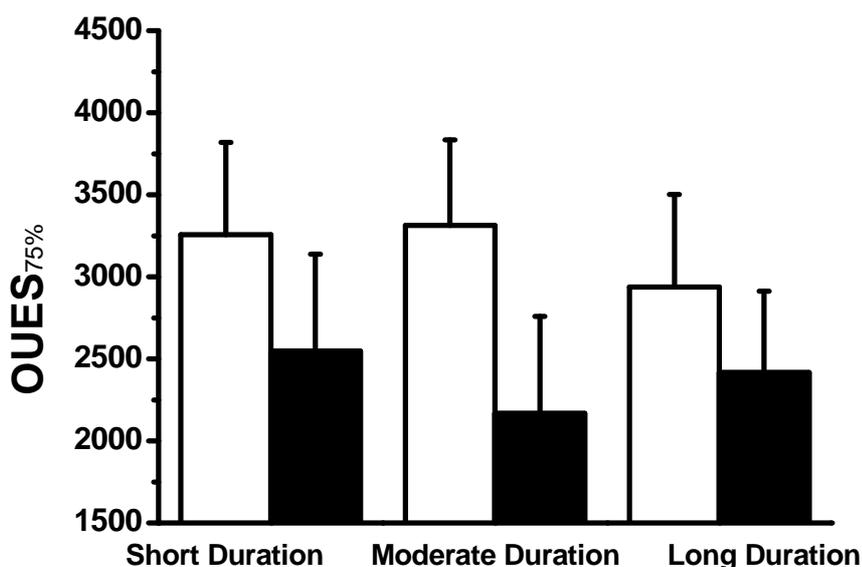


Figure 5-4 illustrates 75% of oxygen uptake efficiency slope OUES_{75%} Total including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.4 shows the impact of short, moderate and long test ramp protocols on the OUES_{75%} determined in the male and female volunteers. Males achieved OUES_{75%} of 3257±563 (range 2367-4042) during the short protocol, 3314±521 (range 2418-4189) during the moderate protocol and 2938±563 (range 1799-3524)

during the long protocol. There was no significant difference in the $OUES_{75\%}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{75\%}$ of 2547 ± 592 (range 1643-3345) during the short protocol, 2167 ± 592 (range 1571-3077) during the moderate protocol and 2417 ± 496 (range 1714-3367) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{75\%}$ achieved by the females in the test ramp protocols. However, the female $OUES_{75\%}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{75\%}$ (excluding cardio-dynamic phase).

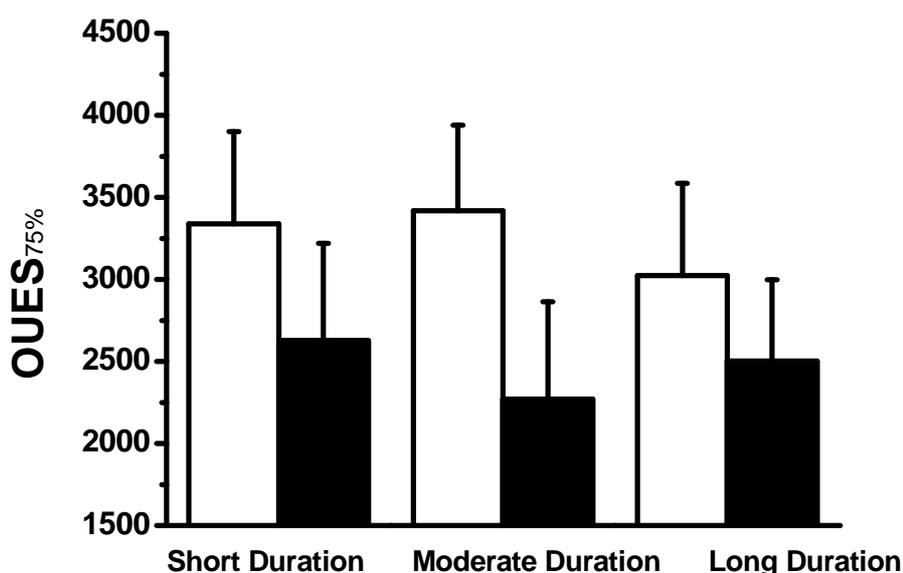


Figure 5-5 illustrates 75% of oxygen uptake efficiency slope $OUES_{75\%}$ Total excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.5 shows the impact of short, moderate and long test ramp protocols on the $OUES_{75\%}$ determined in the male and female volunteers. Males achieved $OUES_{75\%}$ of 3338 ± 563 (range 2448-4123) during the short protocol, 3418 ± 521 (range 2522-4293) during the moderate protocol and 3023 ± 563 (range 1884-3609) during the long protocol. There was no significant difference in the $OUES_{75\%}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{75\%}$ of

2628±592 (range 1724-3426) during the short protocol, 2271±592 (range 1675-3181) during the moderate protocol and 2502±496 (range 1799-3452) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{75\%}$ achieved by the females in the test ramp protocols. However, the female $OUES_{75\%}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{before}$ (including cardio-dynamic phase).

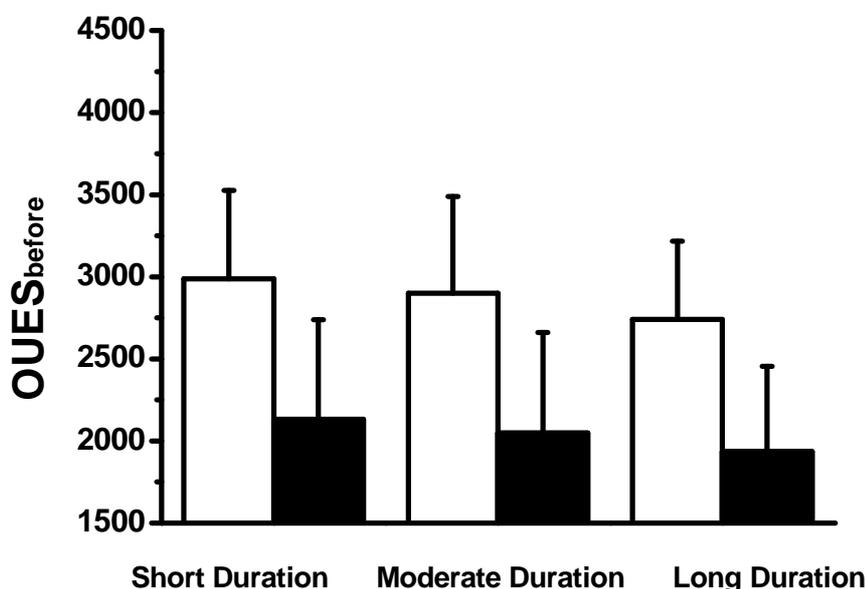


Figure 5-6 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{before}$ including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.6 shows the impact of short, moderate and long test ramp protocols on the $OUES_{before}$ determined in the male and female volunteers. Males achieved $OUES_{before}$ of 2987±539 (range 2239-3750) during the short protocol, 2901±589 (range 1738-3593) during the moderate protocol and 2742±475 (range 1887-3408) during the long protocol. There was no significant difference in the $OUES_{before}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{before}$ of 2131±608 (range 1289-2836) during the short protocol, 2047±615 (range 1240-3036) during the moderate protocol and 1937±517 (range 1146-2938) during the

long protocol. Like their male counterparts, there was no significant difference in the $OUES_{before}$ achieved by the females in the test ramp protocols. However, the female $OUES_{before}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{before}$ (excluding cardio-dynamic phase).

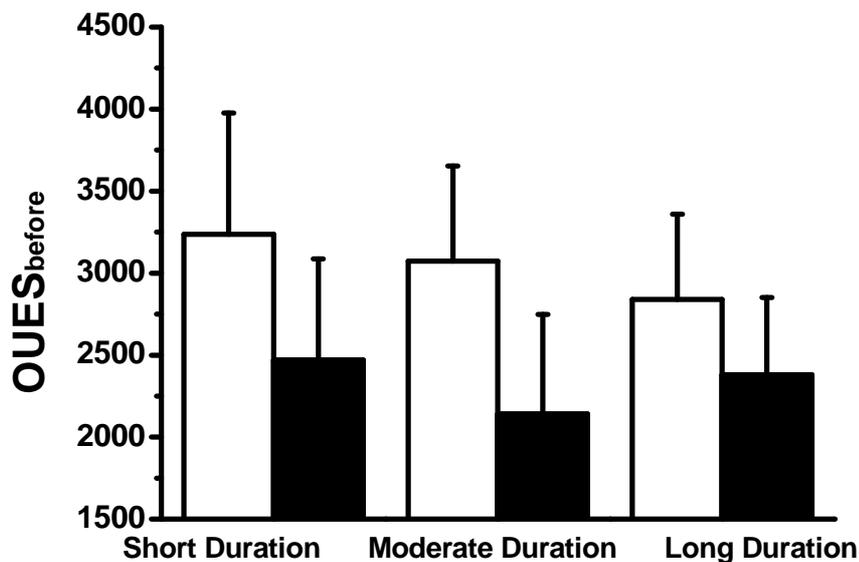


Figure 5-7 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{before}$ excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.7 shows the impact of short, moderate and long test ramp protocols on the $OUES_{before}$ determined in the male and female volunteers. Males achieved $OUES_{before}$ of 3235 ± 741 (range 2211-4606) during the short protocol, 3073 ± 580 (range 2258-3950) during the moderate protocol and 2841 ± 518 (range 1883-3677) during the long protocol. There was no significant difference in the $OUES_{before}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{before}$ of 2470 ± 617 (range 1711-3596) during the short protocol, 2141 ± 607 (range 1266-2874) during the moderate protocol and 2380 ± 471 (range 1770-3197) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{before}$ achieved by the females in the test ramp protocols. However,

the female $OUES_{before}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{after}$ (including cardio-dynamic phase).

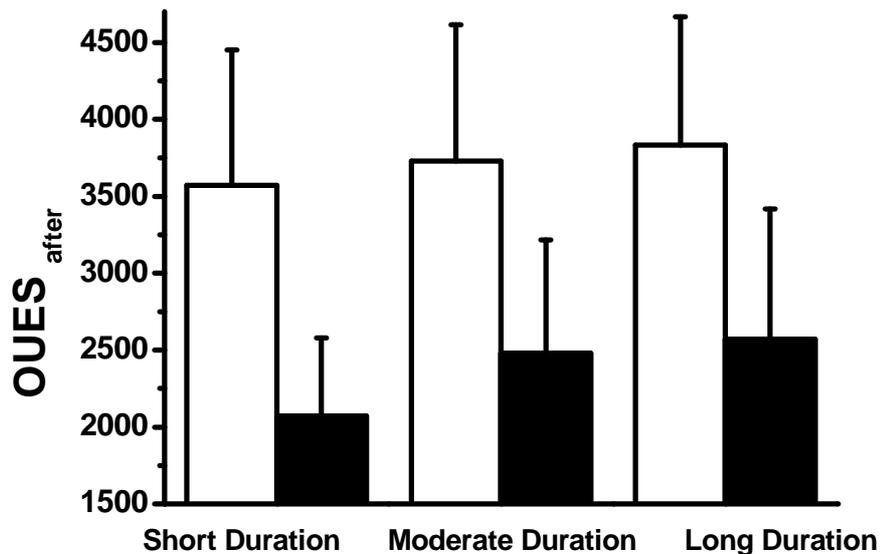


Figure 5-8 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{after}$ including cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.8 shows the impact of short, moderate and long test ramp protocols on the $OUES_{after}$ determined in the male and female volunteers. Males achieved $OUES_{after}$ of 3571 ± 881 (range 2178-4684) during the short protocol, 3729 ± 886 (range 2297-5207) during the moderate protocol and 3833 ± 835 (range 2390-5449) during the long protocol. There was no significant difference in the $OUES_{after}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{after}$ of 2072 ± 508 (range 1418-2858) during the short protocol, 2479 ± 739 (range 1648-3619) during the moderate protocol and 2571 ± 847 (range 1776-4211) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{after}$ achieved by the females in the test ramp protocols. However, the female $OUES_{after}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill $OUES_{after}$ (excluding cardio-dynamic phase).

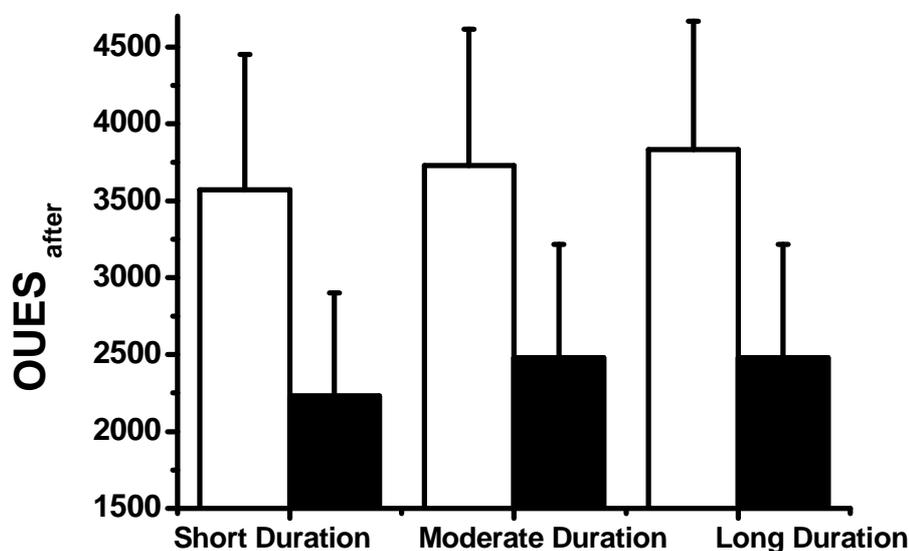


Figure 5-9 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{after}$ excluding cardio-dynamic phase during three treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.9 shows the impact of short, moderate and long test ramp protocols on the $OUES_{after}$ determined in the male and female volunteers. Males achieved $OUES_{after}$ of 3571 ± 881 (range 2178-4684) during the short protocol, 3729 ± 886 (range 2297-5207) during the moderate protocol and 3833 ± 835 (range 2390-5449) during the long protocol. There was no significant difference in the $OUES_{after}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{after}$ of 2230 ± 671 (range 1418-3492) during the short protocol, 2479 ± 739 (range 1648-3619) during the moderate protocol and 2479 ± 739 (range 1648-3619) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{after}$ achieved by the females in the test ramp protocols. However, the female $OUES_{after}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, *t*-test).

Correlation between $\dot{V}_{O_2 \text{ PEAK}}$ and maximal and sub-maximal OUES during moderate duration.

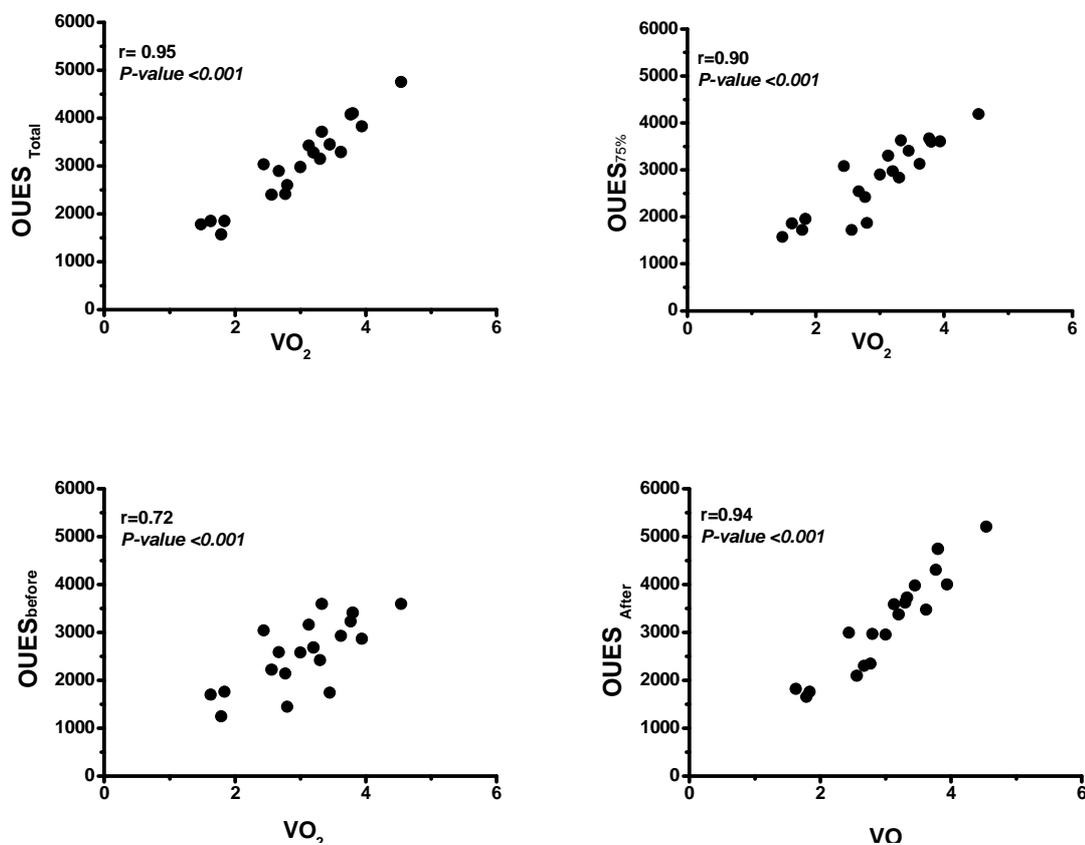


Figure 5-10 Pearson's correlation coefficient measures the linear relations between OUES and \dot{V}_{O_2} (ml/min), during moderate treadmill studies. For both male and female as one group and including cardio-dynamic phase.

Influence of test ramp protocols on cycle ergometer $OUES_{total}$ (including cardio-dynamic phase).

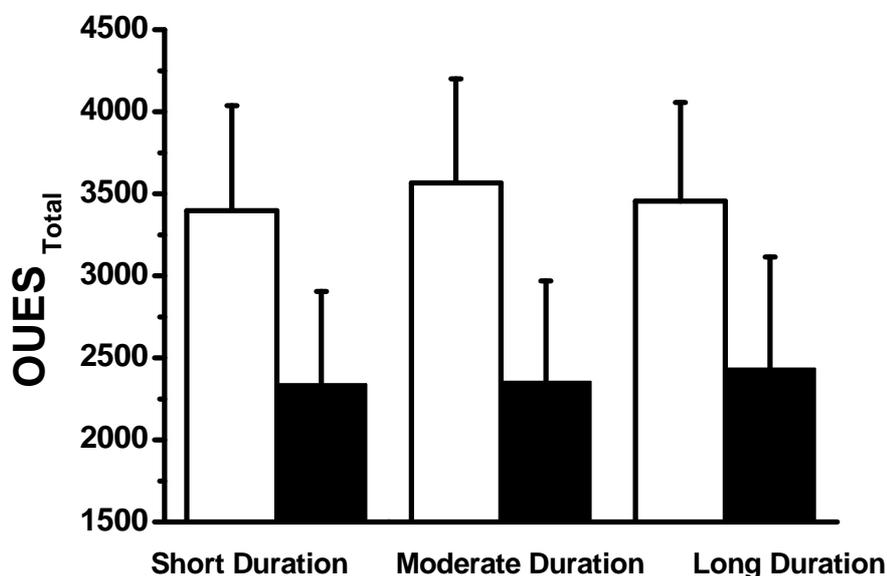


Figure 5-11 illustrates total oxygen uptake efficiency slope $OUES_{Total}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.11 shows the impact of short, moderate and long test ramp protocols on the $OUES_{total}$ determined in the male and female volunteers. Males achieved $OUES_{total}$ of 3398 ± 422 (range 2132-3599) during the short protocol, 3565 ± 615 (range 2264-4189) during the moderate protocol and 3456 ± 516 (range 2485-4132) during the long protocol. There was no significant difference in the $OUES_{total}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{total}$ of 2341 ± 457 (range 1340-2739) during the short protocol, 2356 ± 417 (range 1408-2517) during the moderate protocol and 2436 ± 419 (range 1463-2526) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{total}$ achieved by the females in the test ramp protocols. However, the female $OUES_{total}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on cycle ergometer $OUES_{total}$ (excluding cardio-dynamic phase).

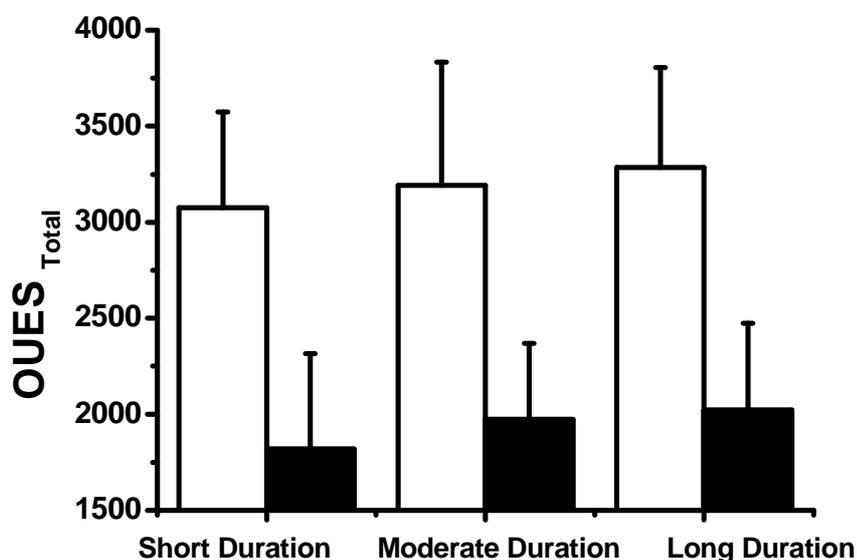


Figure 5-12 illustrates total oxygen uptake efficiency slope $OUES_{Total}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.12 shows the impact of short, moderate and long test ramp protocols on the $OUES_{total}$ determined in the male and female volunteers. Males achieved $OUES_{total}$ of 3077 ± 498 (range 1861-3502) during the short protocol, 3192 ± 643 (range 2153-4209) during the moderate protocol and 3285 ± 520 (range 2664-4310) during the long protocol. There was no significant difference in the $OUES_{total}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{total}$ of 1819 ± 497 (range 1297-2730) during the short protocol, 1975 ± 394 (range 1510-2568) during the moderate protocol and 2022 ± 452 (range 1408-2548) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{total}$ achieved by the females in the test ramp protocols. However, the female $OUES_{total}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on cycle ergometer OUES_{75%} (including cardio-dynamic phase).

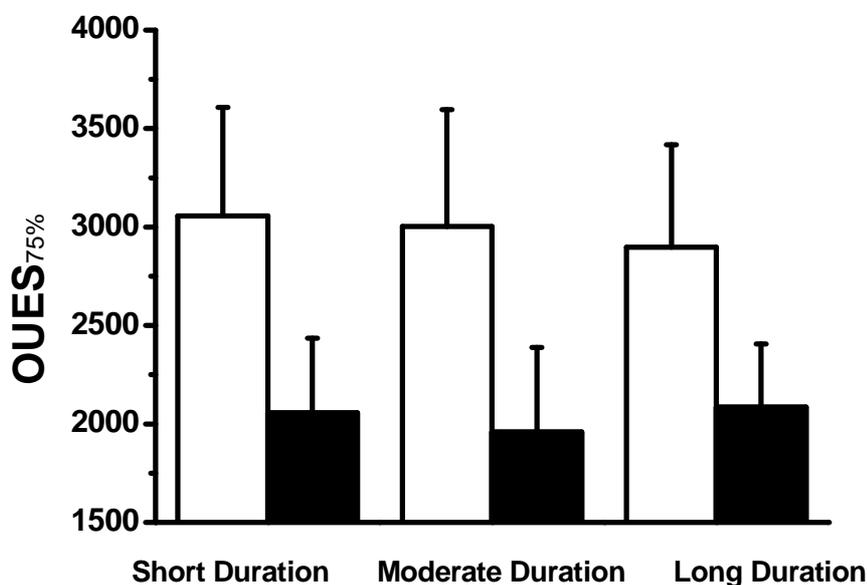


Figure 5-13 illustrates 75% of oxygen uptake efficiency slope OUES_{75%} including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.13 shows the impact of short, moderate and long test ramp protocols on the OUES_{75%} determined in the male and female volunteers. Males achieved OUES_{75%} of 3057±552 (range 2142-3818) during the short protocol, 3003±594 (range 2053-4041) during the moderate protocol and 2898±520 (range 2244-3671) during the long protocol. There was no significant difference in the OUES_{75%} achieved by the males in the test ramp protocols. Females achieved OUES_{75%} of 2057±378 (range 1431-2656) during the short protocol, 1960±429 (range 1474-2455) during the moderate protocol and 2085±322 (range 1583-2506) during the long protocol. Like their male counterparts, there was no significant difference in the OUES_{75%} achieved by the females in the test ramp protocols. However, the female OUES_{75%} was significantly lower than the male during all test ramp protocols ($P < 0.05$, *t*-test).

Influence of test ramp protocols on cycle ergometer OUES_{75%} (excluding cardio-dynamic phase).

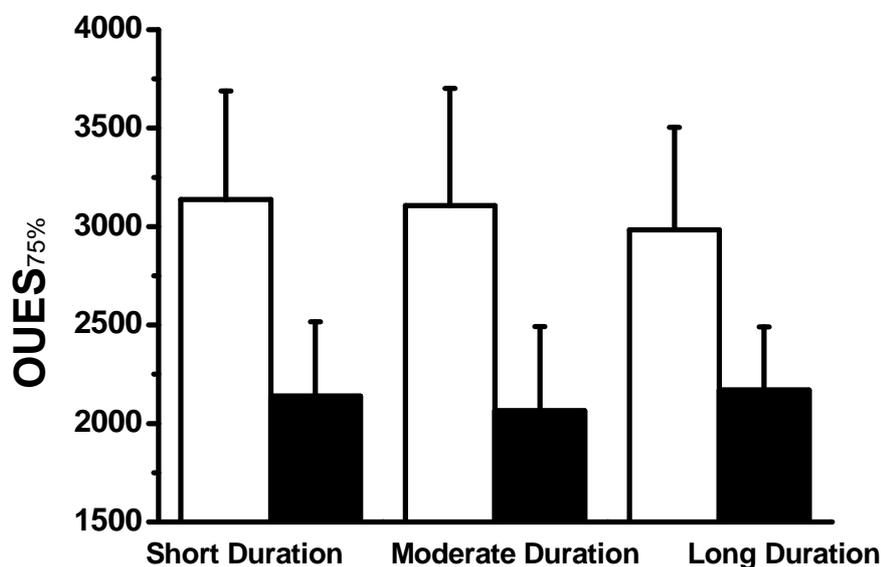


Figure 5-14 illustrates 75% of oxygen uptake efficiency slope OUES_{75%} excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.14 shows the impact of short, moderate and long test ramp protocols on the OUES_{75%} determined in the male and female volunteers. Males achieved OUES_{75%} of 3138±552 (range 2223-3899) during the short protocol, 3107±594 (range 2157-4145) during the moderate protocol and 2983±520 (range 2329-3756) during the long protocol. There was no significant difference in the OUES_{75%} achieved by the males in the test ramp protocols. Females achieved OUES_{75%} of 2138±378 (range 1512-2737) during the short protocol, 2064±429 (range 1578-2559) during the moderate protocol and 2170±322 (range 1668-2591) during the long protocol. Like their male counterparts, there was no significant difference in the OUES_{75%} achieved by the females in the test ramp protocols. However, the female OUES_{75%} was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on cycle ergometer $OUES_{before}$ (including cardio-dynamic phase).

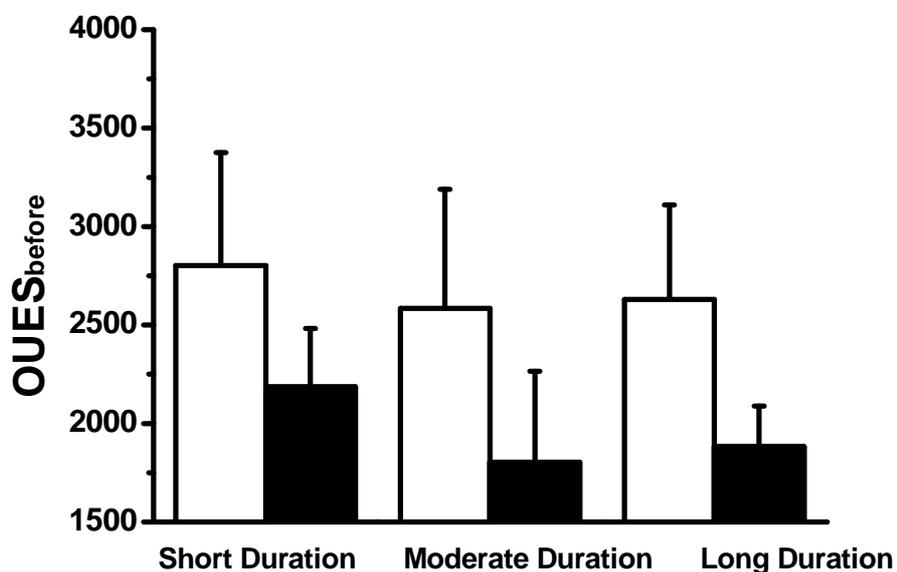


Figure 5-15 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{before}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.15 shows the impact of short, moderate and long test ramp protocols on the $OUES_{before}$ determined in the male and female volunteers. Males achieved $OUES_{before}$ of 2802 ± 575 (range 1918-3731) during the short protocol, 2585 ± 606 (range 1646-3769) during the moderate protocol and 2631 ± 480 (range 1746-3162) during the long protocol. There was no significant difference in the $OUES_{before}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{before}$ of 2186 ± 296 (range 1796-2698) during the short protocol, 1803 ± 463 (range 1050-2404) during the moderate protocol and 1883 ± 205 (range 1585-2065) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{before}$ achieved by the females in the test ramp protocols. However, the female $OUES_{before}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on cycle ergometer $OUES_{before}$ (excluding cardio-dynamic phase).

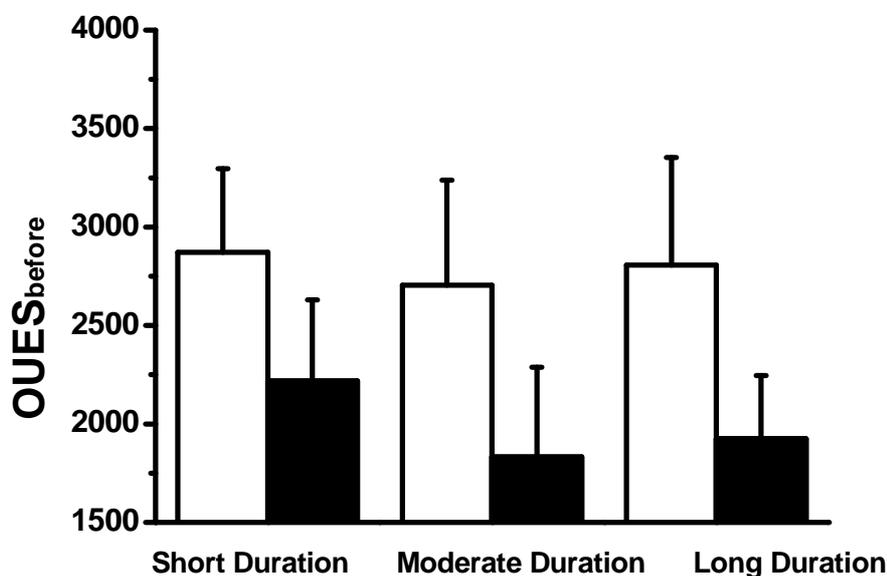


Figure 5-16 illustrates oxygen uptake efficiency slope before anaerobic threshold brake point $OUES_{before}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.16 shows the impact of short, moderate and long test ramp protocols on the $OUES_{before}$ determined in the male and female volunteers. Males achieved $OUES_{before}$ of 2873 ± 424 (range 2172-3447) during the short protocol, 2704 ± 535 (range 1800-3613) during the moderate protocol and 2808 ± 545 (range 1741-3404) during the long protocol. There was no significant difference in the $OUES_{before}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{before}$ of 2218 ± 413 (range 1737-2848) during the short protocol, 1833 ± 454 (range 993-2404) during the moderate protocol and 1925 ± 321 (range 1435-2555) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{before}$ achieved by the females in the test ramp protocols. However, the female $OUES_{before}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, *t*-test).

Influence of test ramp protocols on cycle ergometer $OUES_{after}$ (including cardio-dynamic phase).

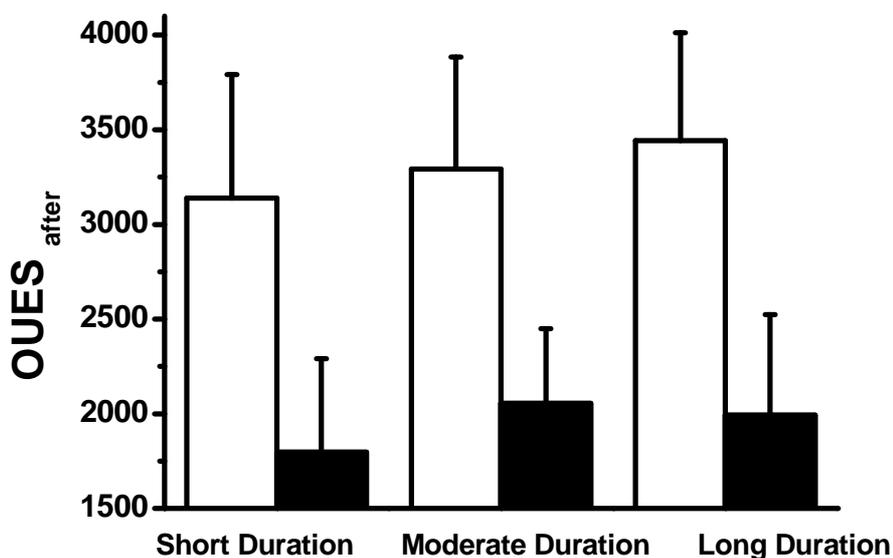


Figure 5-17 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{after}$ including cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.17 shows the impact of short, moderate and long test ramp protocols on the $OUES_{after}$ determined in the male and female volunteers. Males achieved $OUES_{after}$ of 3140 ± 651 (range 1871-3879) during the short protocol, 3293 ± 592 (range 2197-4152) during the moderate protocol and 3443 ± 570 (range 2746-4663) during the long protocol. There was no significant difference in the $OUES_{after}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{after}$ of 1796 ± 495 (range 1265-2682) during the short protocol, 2055 ± 394 (range 1567-2697) during the moderate protocol and 1993 ± 531 (range 1276-2564) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{after}$ achieved by the females in the test ramp protocols. However, the female $OUES_{after}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on cycle ergometer $OUES_{after}$ (excluding cardio-dynamic phase).

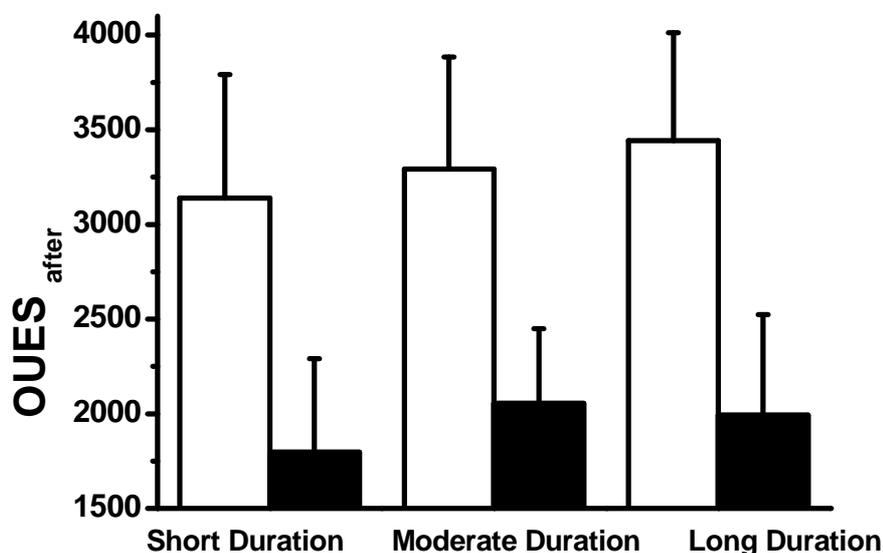


Figure 5-18 illustrates oxygen uptake efficiency slope after anaerobic threshold brake point $OUES_{after}$ excluding cardio-dynamic phase during three cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 5.18 shows the impact of short, moderate and long test ramp protocols on the $OUES_{after}$ determined in the male and female volunteers. Males achieved $OUES_{after}$ of 3140 ± 651 (range 1871-3879) during the short protocol, 3293 ± 592 (range 2197-4152) during the moderate protocol and 3443 ± 570 (range 2746-4663) during the long protocol. There was no significant difference in the $OUES_{after}$ achieved by the males in the test ramp protocols. Females achieved $OUES_{after}$ of 1796 ± 495 (range 1265-2682) during the short protocol, 2055 ± 394 (range 1567-2697) during the moderate protocol and 1993 ± 531 (range 1276-2564) during the long protocol. Like their male counterparts, there was no significant difference in the $OUES_{after}$ achieved by the females in the test ramp protocols. However, the female $OUES_{after}$ was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Correlation between $\dot{V}_{O_2 \text{ PEAK}}$ and maximal and sub-maximal OUES during moderate duration.

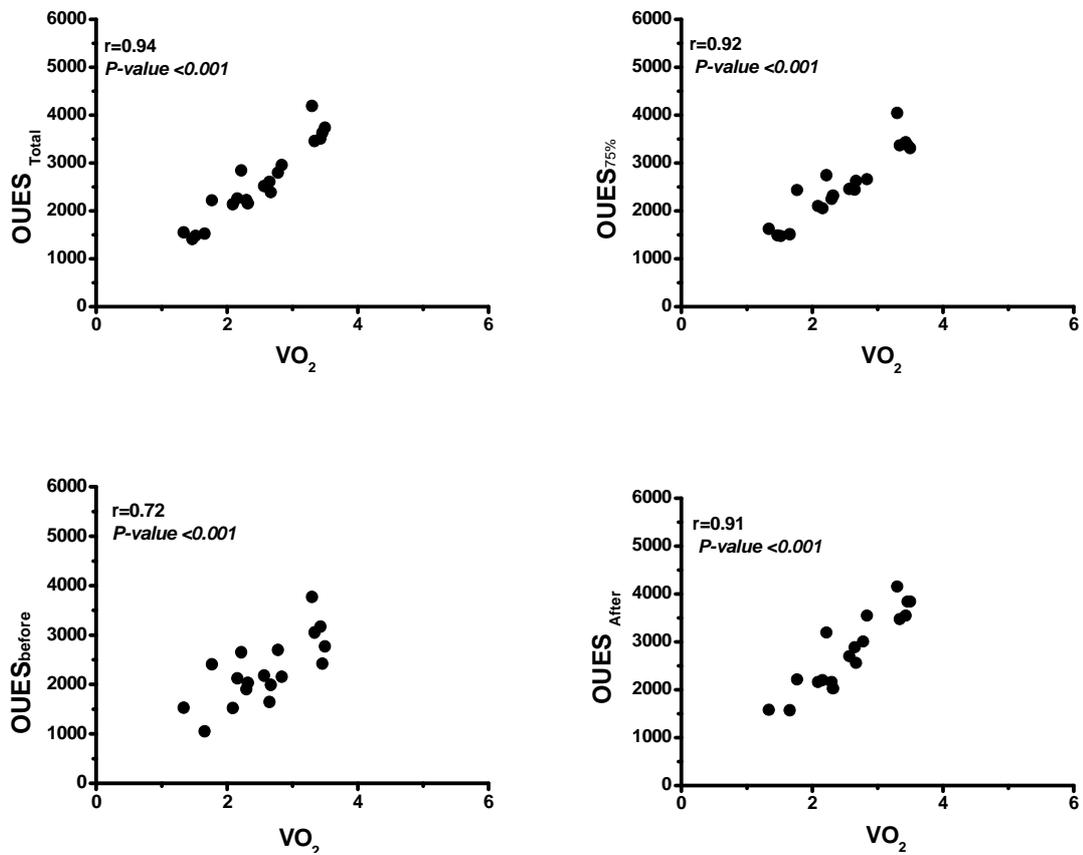


Figure 5-19 Pearson's correlation coefficient measures the linear relations between OUES and \dot{V}_{O_2} (ml/min), during moderate cycle ergometer studies. For both male and female as one group and including cardio-dynamic phase.

5.7 Discussion

The main aim of the current study was to determine whether exercise duration influenced the OUES measured during a ramp exercise protocol and as with the previous chapters it is evident that the measurement is independent of exercise duration and exercise modality.

These results support the previous data from Baba *et al* (1999) who found that found that short and long duration treadmill exercise did not influence the OUES (Baba *et al.*, 1996) However, these data also show that the OUES can be measured independent of exercise modality and again that it is not influenced by duration on a cycle. The fact that the measurement is modality and duration independent will allow the subsequent discussion of the relative proportions of data analysed from a (CPET) session to be discussed in general terms.

During the first couple of minutes of steady-state exercise, the relationship between ventilation and oxygen uptake is complex due to the non-aerobic production of ATP from immediate energy stores and anaerobic glycolysis. For this reason, it is often recommended that the the first 2-3 minutes of breath by breath data collection is removed from subsequent data analysis (Balady *et al.*, 2010). The standard analysis of OUES does not remove this data and, consequently, this study also investigated the impact of the 'so called' cardio-dynamic phase on the calculation of the OUES.

In the current study, removing the first 2 minutes from breath by breath data before data analysis had little effect on the subsequently derived OUES. This is, perhaps, unsurprising because of the log transformation applied to the data to determine a linear slope from non-linear data (Van Laethem *et al.*, 2006). Conventional wisdom dictates that data should be analysed untransformed and that residuals should be examined for outliers and deviations from Normality. The inclusion of the cardio-dynamic phase introduces an easily modified departure from the required assumptions for linear analysis that can be applied to determine whether the effects of the log transformation are appropriate. The fact that exclusion of the cardio-dynamic phase in a heterogeneous groups of volunteers had no significant effect upon the measurement of OUES is, therefore, a further validation of the measurement process proposed by Baba *et*

al (1996) for the determination of the OUES. This is important due to the previous observations that an impaired cardio-dynamic phase can significantly influence phase II oxygen uptake kinetics (M'Bouh *et al*, 2001). It remains to be seen whether a similar effect is apparent in diseased populations.

The majority of previous studies have been conducted on frail individuals, in children and in the elderly. In such populations, there is significant benefit in avoiding maximal testing due to the risks from high intensity exercise (Arena *et al.*, 2009). Consequently, some effort has been expended on determining whether restricting the proportion of exercise analysed to determine the OUES has an effect on the measurement obtained. A study by Pogliaghi *et al* calculated OUES from 75%, 90% and 100% of the exercise test duration during cycle ergometer exercise in healthy aged volunteers (Pogliaghi *et al.*, 2007). The study showed no significant difference in OUES measured during maximal and sub-maximal exercise. A similar observation has been made in young children during cycle ergometry (Akkerman *et al.*, 2010).

These observations are of further interest to the validity, specificity and sensitivity of the OUES measurement in clinical practice and form the basis for the second aim of the current study; to determine the influence of restricted data analysis on OUES determination. However, rather than just restricting the analysis to a defined proportion of the exercise data this study has additionally analysed the data in relation to the exercise intensity domain (*i.e.* before and after the lactate threshold or more accurately the ventilatory threshold in this instance).

As with previous studies, the current investigation found that there was no significant difference in the OUES measured from the whole exercise data or when restricted to the first 75% of the exercise data. Furthermore, there was no significant difference in the OUES measured before or after the 'lactate threshold' as determined by the ventilatory threshold (see chapter 4). However, there was a tendency for the OUES to be lower when data analysis was restricted to the data before the threshold and this may impact on the specificity and sensitivity of the OUES for predicting aerobic capacity.

The clinical relevance of OUES is mainly to reflect functional capacity in frail and limited patient populations. Previous studies have demonstrated that the OUES correlates fairly well with aerobic capacity $\dot{V}_{O_2 \text{ PEAK}}$ in a wide range of populations (Hollenberg & Tager, 2000; Baba *et al.*, 1996; Baba *et al.*, 1999; Van Laethem *et al.*, 2006). However, Pichon *et al.* (2002) reported that there were inter-individual variations between $\dot{V}_{O_2 \text{ MAX}}$ and OUES despite a significant correlation and have suggested that OUES might be limited in clinical practice (Pichon *et al.*, 2002). The tendency for a lower OUES to be measured when analysis is restricted below the 'lactate' threshold in this study may go some way to suggesting a reason for this relatively poor sensitivity.

The $\dot{V}_{O_2 \text{ PEAK}}$ is a reliable marker of functional capacity but more accurately it is a marker of aerobic capacity, whereas the lactate threshold is a function of anaerobic capacity. The lactate threshold is not set at a precise proportion of $\dot{V}_{O_2 \text{ PEAK}}$ and consequently if it is at or around, for example, 75% of total exercise then the duration then the OUES will reflect analysis before the lactate threshold if the analysis is restricted to 75% of total exercise duration. Whereas, in less anaerobically 'fit' patients the lactate threshold may occur significantly before 75% of the total exercise duration and reflect more of the total exercise duration response even when restricted to a proportion of total exercise duration.

This is not likely to affect the specificity of the measurement for predicting functional capacity (e.g. NYHA functional capacity) but may limit the capacity to sensitively predict the $\dot{V}_{O_2 \text{ PEAK}}$. This data is similar to the results of removing the cardio-dynamic phase described previously, in that it validates the use of Baba's log transformation for measuring the OUES. The process allows functional classification of frail and diseased population but by diluting (10-fold) the variability of the ventilatory response at higher exercise intensities it will lead to a reduction in sensitivity.

However, this interpretation is purely speculative because the data from the current study do not support a statistically significant difference in the OUES below the lactate threshold. A single previous study, by Van Laethem *et al.* (2009) did try to determine differences in OUES above and below the lactate

threshold but found it difficult to accurately identify the ventilatory threshold in one quarter of their volunteers and were, therefore, unable to show significant differences. It may be prudent to conduct further such studies in diseased populations and using the approach detailed in chapter 4 to allow accurate determination of the ventilatory threshold and subsequently to determine the effects of exercise intensity domains on the measurement of OUES.

CHAPTER SIX

Peak Breathing Reserve Index

6 Impact of Exercise Duration on Peak Breathing Reserve Index and Breathing Reserve Index at Anaerobic Threshold

6.1 Introduction

Breathlessness or dyspnoea is a relatively common symptom that limits exercise capacity in patients undergoing cardio-pulmonary exercise testing (CPET). In some cases, the CPET is terminated due to the patient suffering from breathing discomfort that occurs before the expected criteria of maximal exercise testing is achieved *e.g.* reaching 85% of predicted HR_{max} or 70% of predicted maximum ventilation (Mahler & Franco, 1996). These observations have led to a number of investigators studying the proportion of maximum available ventilation that can be recruited as a measurement of functional capacity. These investigations have led to the use of the breathing reserve index at peak exercise (BRI_{peak}) and the breathing reserve index at anaerobic threshold (BRI_{AT}) to predict the functional capacity (Medoff *et al.*, 1998; Ross, 2003; Sexauer *et al.*, 2003; Tantisira *et al.*, 2002).

The (BRI_{AT}) is a sub-maximal marker of functional capacity that may be useful in patients with limited pulmonary function that could be at risk from maximal exercise testing. The technique has been used to discriminate between Cystic Fibrosis patients with ventilatory and non-ventilatory exercise limitations. Furthermore, the (BRI_{AT}) was shown to correlate well with (BRI_{peak}) ($r = 0.89$; $p < 0.01$) in this study and provides confidence that this sub-maximal marker can be used to assess the pulmonary mechanical limitation in patients with poor functional capacity (Sexauer *et al.*, 2003). These data are supported by a further study in chronic obstructive pulmonary disease (COPD) patients where (BRI_{AT}) correlated well with (BRI_{peak}) ($r=0.85$, $p<0.0001$). In this study, the BRI_{AT} was able to distinguish between pulmonary mechanical and cardiovascular limitations to exercise (Medoff *et al.*, 1998). In addition to being a useful marker of functional capacity, (BRI_{AT}) is a good prognostic marker in lung

disease. Tantisira *et al* (2002) demonstrated that having $(BRI_{AT}) \geq 0.70$ predicted mortality in Cystic Fibrosis patients awaiting lung transplantation (Tantisira *et al.*, 2002).

The breathing reserve is simply the difference between the ventilation at a given point and the maximum ventilation available to that individual. Obviously, the potential limiting factor to this marker of functional capacity is in the accurate description of the maximum ventilation available to that individual. The 12-s maximum voluntary ventilation (MVV) is commonly employed as a marker of the maximum ventilation available to an individual and correlates well with peak ventilation during maximal exercise testing (Dillard *et al.*, 1993). This data is further supported by a comprehensive study to assess the relationship of (MVV) with peak ventilation. In this study, there was no statistical difference between the (MVV) and peak ventilation during exercise in 231 healthy volunteers (Blackie *et al.*, 1991). A breathing reserve of less than $11 \text{ l}\cdot\text{min}^{-1}$ is considered abnormal (Hansen *et al.*, 1984).

However, the technique of measuring (MVV) is difficult and can be problematic in patient populations ((Kor *et al.*, 2004). For this reason, most clinical studies estimate maximum voluntary ventilation from the forced expiratory volume in 1 second (FEV_1) (Gandevia & Hugh-Jones, 1957; Miller *et al.*, 1959; Kor *et al.*, 2004) but maybe less accurate than direct measurement (Dillard *et al.*, 1993). Normally, the breathing frequency during (MVV) measurement is around 70-110 breaths per minutes and requires deep inhalation and expiration for 12 to 15 seconds. This manoeuvre requires the ability to calls upon components of the pulmonary system to support good tidal volumes, breathing frequency and lung expansion (Morris, 1976). Since the prediction from FEV_1 does not directly assess limits on lung expansion during high frequency breathing it can overestimate (MVV) in patient populations.

The previous discussion in chapters three and four highlight that exercise duration does not influence peak ventilation or the ventilatory threshold measured during ramp exercise protocols. From this, we could presume that there will be no effect of exercise duration on markers of breathing reserve. However, non-significant differences in measuring ventilation and ventilatory

threshold may be additive and result in a significant difference in breathing reserve. In the current study, the (MVV) was measured directly to ensure that the measurement was as accurate as possible to facilitate an assessment of the effects of exercise duration on (BRI_{peak}) and (BRI_{AT}). In addition, the ventilatory threshold was determined using well established procedures to ensure the validity of the measurement.

Moreover, in the main, (BRI_{peak}) and (BRI_{AT}) measurements have been obtained during cycle ergometer exercise and this study has included measurements from treadmill exercise. The physiological demand of treadmill exercise is greater and results in higher peak ventilation that will reduce the breathing reserve. These data will also allow the influence of exercise modality on breathing reserve to be assessed.

6.2 Method

Subjects

Analysis was restricted to data from volunteers that completed three treadmill or three cycle ergometer studies. The volunteer characteristics are summarised in Table 6.1.

Variable	Gender	Treadmill	Cycle Ergometer
Number of Participants	Male	11	11
	Female	9	9
Age (years)	Male	31.2±6.0 (24-42)	32.5±6.0 (25-42)
	Female	36.6±11.0 * (24-57)	35.4±10.0 * (24-57)
Weight (kg)	Male	76.0±7.7 (60-84)	79.0±12.5 (60-103)
	Female	60.7±9.0 * (48-77)	60.0±9.0 * (48-77)
Height (cm)	Male	175.0±6.4 (163-182)	175.4±5.7 (163-182)
	Female	162.0±9.0 * (146-175)	160.7±7.3 * (145-172)
BSA [†] (m) ²	Male	1.92±0.14 (1.94-2.02)	1.96±0.12 (1.65-2.22)
	Female	1.65±0.14 * (1.47-1.87)	1.64±0.13 * (1.47-1.87)
BMI [#] (kg.m ²)	Male	24.8±2.0 (21-28)	26.0±4.0 (21-34)
	Female	23.1±3.8 * (18-29)	23.0±4.0 * (18-29)

Table 6.1 Physical characteristics of all study volunteers. Data are presented as mean±SD (where appropriate) and the range is given in parenthesis. * indicates $P<0.05$ compared with males (t -test with Bonferroni correction). [†] BSA is Body Surface Area according to Mosteller formula. [#] BMI is Body Mass Index.

6.3 Calculation of breathing reserve index

Breathing reserve index (BRI) was calculated at peak exercise (BRI_{peak}) and at the ventilatory threshold (BRI_{AT}). The relative fraction of peak exercise ventilation ($\dot{V}_{E_{PEAK}}$) to maximal voluntary ventilation (MVV) is (BRI_{peak}). The relative fraction of ventilation at the ventilatory threshold (see chapter 4 for details) to

maximal voluntary ventilation (MVV) is (BRI_{AT}) (Medoff *et al.*, 1998; Ross, 2003; Sexauer *et al.*, 2003; Tantisira *et al.*, 2002).

Maximum voluntary ventilation (MVV) was measured over a 12-second period at rest using an open-circuit spirometer (Jaeger® Oxycon Pro®, Viasys health care, Version 5.2). The spirometer was calibrated before each test according to the manufacturer's instructions (Jaeger® Oxycon Pro®, instruction manual 2007).

Volunteers performed two trial measurements of (MVV) and the highest measured value was used for subsequent calculations.

6.4 Statistical analysis

All data are presented as mean \pm standard deviation (SD) and all statistical analyses were performed using Origin Pro 8 SR4 v8. 0951(B951) and SPSS (18.0.0. Chicago). At the 0.05 level, all sub-maximal variables are normally distributed by Shapiro-Wilks normality test and suitable for parametric data analysis. All (BRI_{Peak}) and (BRI_{AT}) in both exercise modalities were significantly drawn from a normal distributed population. A Repeated measurement ANOVA was used to examine the differences between (BRI_{Peak}) and (BRI_{AT}) during short, moderate and long exercise duration studies and this finding was obtained by analysed the data of subjects as whole or according to the gender differences. If a significant was obtained, a *post hoc* test was used for multiple comparisons between means with significance taken as $P < 0.05$. Pearson's correlation coefficients were used to establish the relationship between (BRI_{Peak}) and (BRI_{AT}).

6.5 Results

Physical characteristics

Table 6.1 indicates that the study participants are heterogeneous showing a wide range in age and body habitus. However, there was no significant difference in the physical characteristics between the two exercise modality groups. As expected, the male volunteers were significantly taller and heavier (resulting in higher BSA and BMI) than the female volunteers. The female group were significantly older than the males.

6.6 Influence of test ramp protocols on exercise duration

The different biomechanics and energy demands of walking and cycling can influence cardiopulmonary measurements during maximal effort. Consequently, the data for each exercise modality are presented separately.

Influence of test ramp protocols on treadmill (BRI_{Peak})

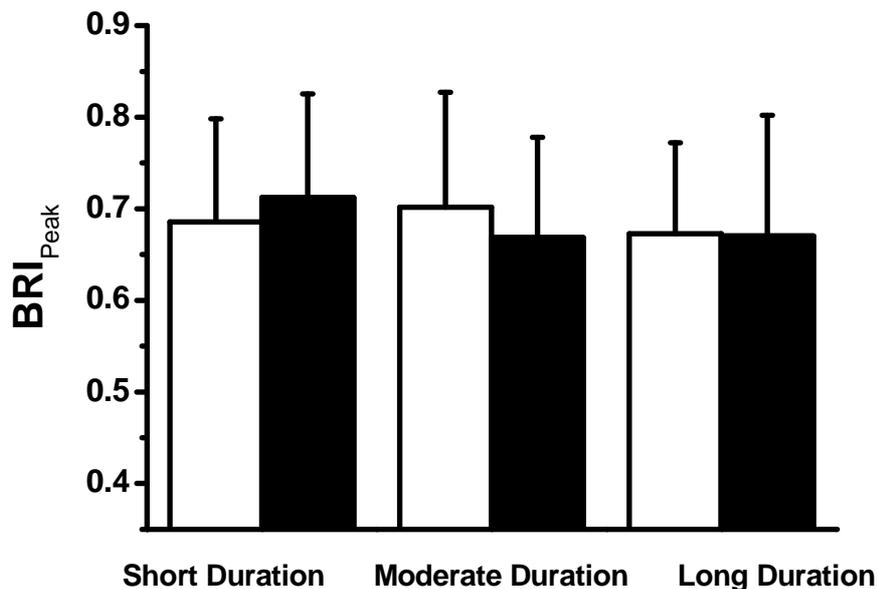


Figure 6-1 illustrates peak breathing reserve index (BRI_{Peak}) during short moderate and long treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6.1 shows the impact of short, moderate and long test ramp protocols on the (BRI_{Peak}) determined in the male and female volunteers. Males achieved (BRI_{Peak}) of 0.69 ± 0.11 (range 0.54-0.86) during the short protocol, 0.70 ± 0.13 (range 0.49-0.87) during the moderate protocol and 0.67 ± 0.1 (range 0.56-0.85) during the long protocol. There was no significant difference in the (BRI_{Peak}) achieved by the males in the test ramp protocols. Females achieved (BRI_{Peak}) of 0.71 ± 0.11 (range 0.52-0.86) during the short protocol, 0.67 ± 0.11 (range 0.47-0.82) during the moderate protocol and 0.67 ± 0.13 (range 0.44-0.82) during the long protocol. Like their male counterparts, there was no significant difference in the (BRI_{Peak}) achieved by the females in the test ramp protocols. However,

the female (BRI_{Peak}) was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Influence of test ramp protocols on treadmill (BRI_{AT})

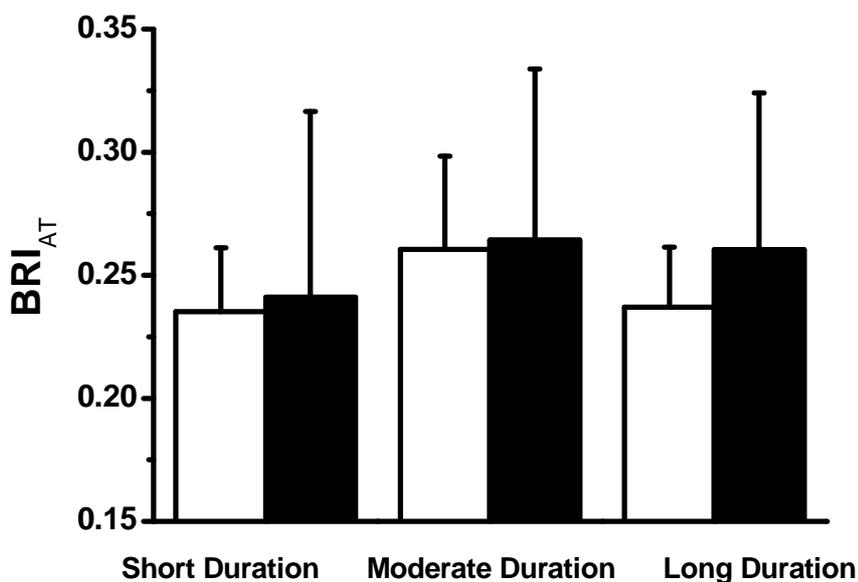


Figure 6-2 illustrates peak breathing reserve index at anaerobic threshold point (BRI_{AT}) during short moderate and long treadmill studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6.2 shows the impact of short, moderate and long test ramp protocols on the BRI_{AT} determined in the male and female volunteers. Males achieved BRI_{AT} of 0.24 ± 0.03 (range 0.20-0.27) during the short protocol, 0.26 ± 0.04 , (range 0.20-0.31) during the moderate protocol and 0.24 ± 0.02 (range 0.20-0.27) during the long protocol. There was no significant difference in the BRI_{AT} achieved by the males in the test ramp protocols. Females achieved BRI_{AT} of 0.24 ± 0.1 (range 0.16-0.39) during the short protocol, 0.26 ± 0.1 (range 0.17-0.39) during the moderate protocol and 0.26 ± 0.1 (range 0.18-0.34) during the long protocol. Like their male counterparts, there was no significant difference in the BRI_{AT} achieved by the females in the test ramp protocols. However, the female BRI_{AT} was significantly lower than the male during all test ramp protocols ($P < 0.05$, t -test).

Correlation between (BRI_{Peak}) and (BRI_{AT}) during treadmill studies

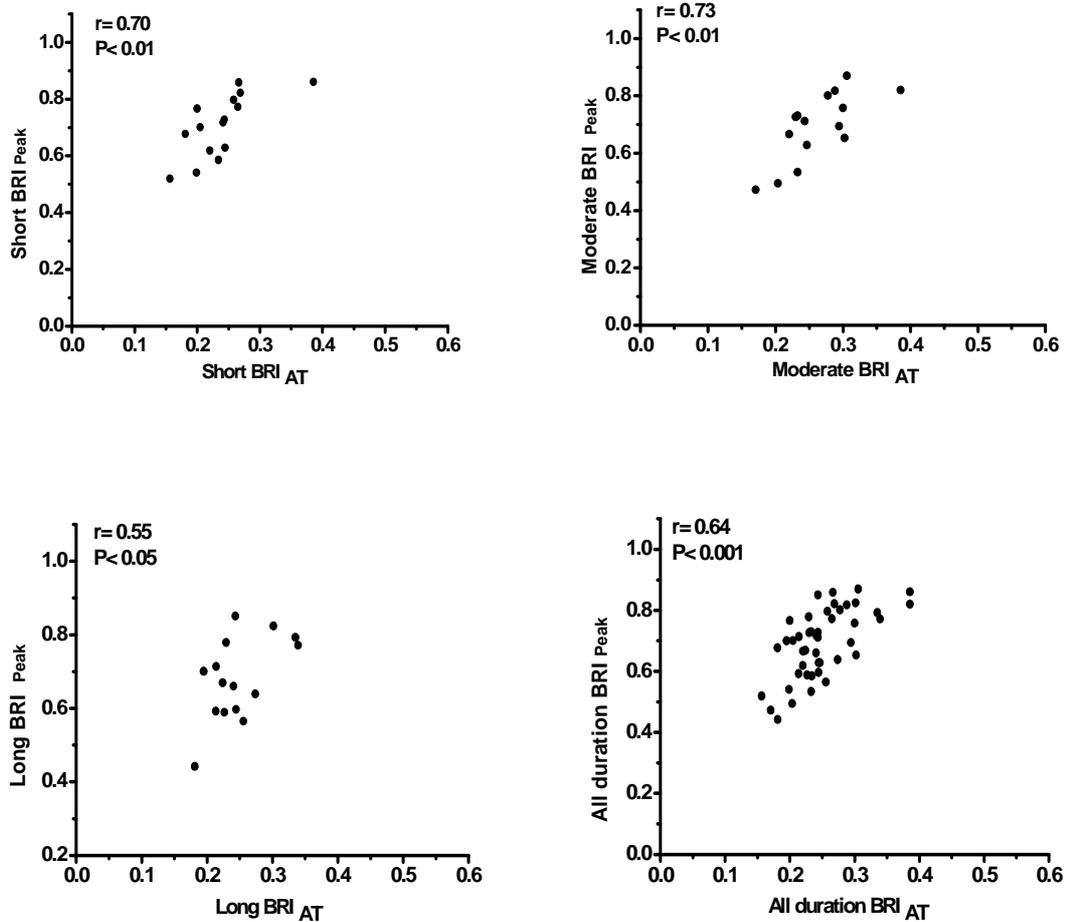


Figure 6-3 Correlation between (BRI_{Peak}) and (BRI_{AT}) during short, moderate and long exercise duration studies and for all data together during treadmill studies.

Figure 6.3 demonstrates that there is a good agreement between (BRI_{Peak}) and (BRI_{AT}) during the test ramp protocols on the treadmill.

Influence of test ramp protocols on cycle ergometer (BRI_{Peak})

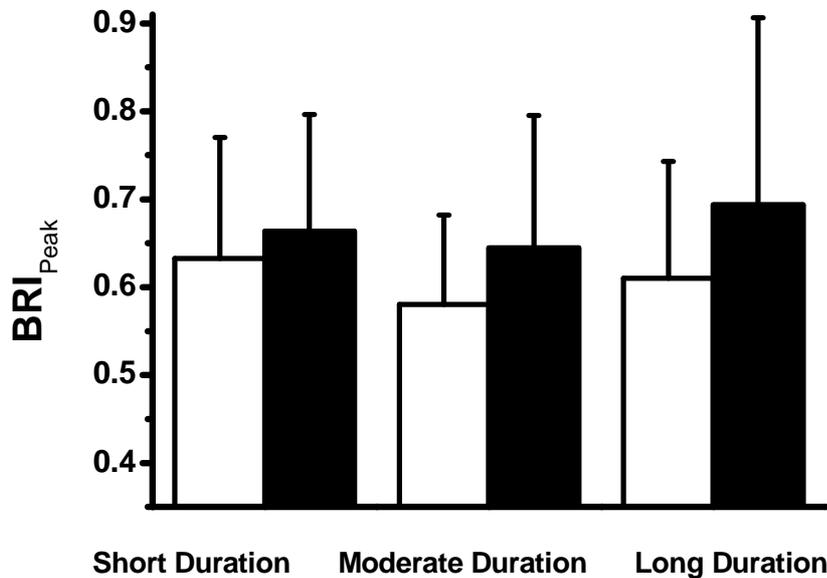


Figure 6-4 illustrates peak breathing reserve index (BRI_{Peak}) during short moderate and long cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6.4 shows the impact of short, moderate and long test ramp protocols on the (BRI_{Peak}) determined in the male and female volunteers. Males achieved (BRI_{Peak}) of 0.63 ± 0.14 (range 0.48-0.84) during the short protocol, 0.58 ± 0.10 (range 0.48- 0.77) during the moderate protocol and 0.61 ± 0.13 (range 0.42-0.86) during the long protocol. There was no significant difference in the (BRI_{Peak}) achieved by the males in the test ramp protocols. Females achieved (BRI_{Peak}) of 0.66 ± 0.13 (range 0.49-0.90) during the short protocol, 0.64 ± 0.15 (range 0.45-0.84) during the moderate protocol and 0.69 ± 0.21 (range 0.39-1) during the long protocol. Like their male counterparts, there was no significant difference in the (BRI_{Peak}) achieved by the females in the test ramp protocols. However, the female (BRI_{Peak}) was significantly lower than the male during all test ramp protocols ($P < 0.05$, *t*-test).

Influence of test ramp protocols on cycle ergometer BRI_{AT}

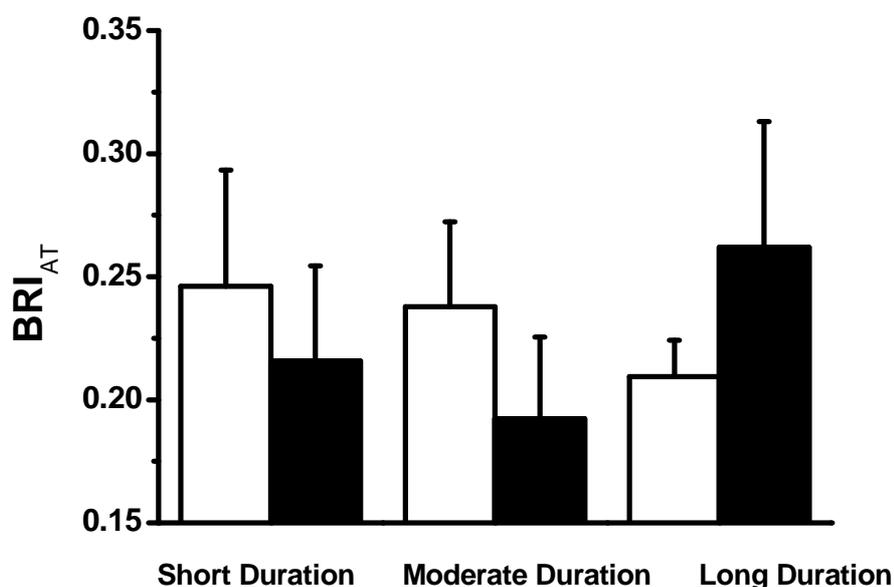


Figure 6-5 illustrates peak breathing reserve index at anaerobic threshold point (BRI_{AT}) during short moderate and long cycle ergometer studies. Male volunteers are represented by the open bars and female volunteers by the closed bars.

Figure 6.5 shows the impact of short, moderate and long test ramp protocols on the (BRI_{AT}) determined in the male and female volunteers. Males achieved (BRI_{AT}) of 0.25 ± 0.04 (range 0.19-0.23) during the short protocol, 0.24 ± 0.03 (range 0.18-0.28) during the moderate protocol and 0.21 ± 0.01 (range 0.16-0.31) during the long protocol. There was no significant difference in the (BRI_{AT}) achieved by the males in the test ramp protocols. Females achieved (BRI_{AT}) of 0.22 ± 0.04 , range 0.21-0.34 during the short protocol, 0.19 ± 0.03 , range 0.13-0.23 during the moderate protocol and 0.26 ± 0.05 , range 0.16-0.26 during the long protocol. Like their male counterparts, there was no significant difference in the BRI_{AT} achieved by the females in the test ramp protocols. However, the female BRI_{AT} was significantly lower than the male during all test ramp protocols ($P < 0.05$, *t*-test).

Correlation between (BRI_{Peak}) and (BRI_{AT}) during cycle ergometer studies.

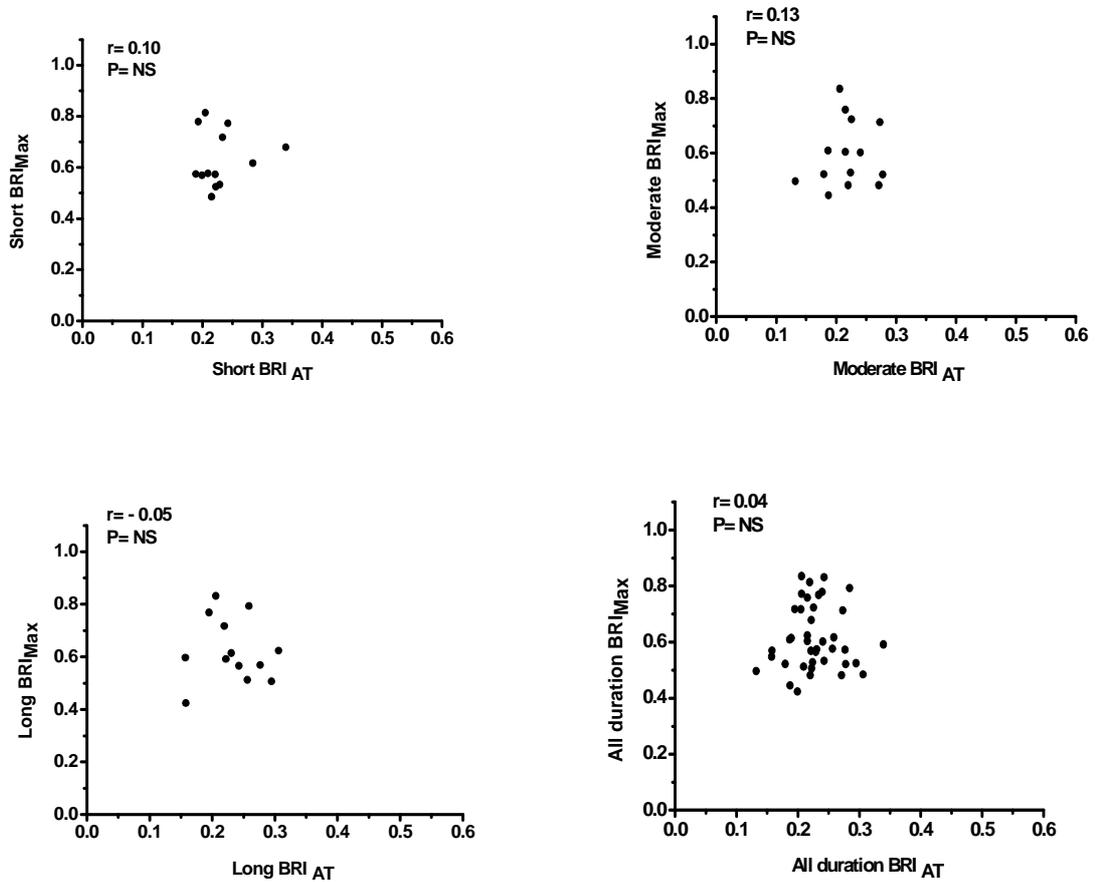


Figure 6-6 Correlation between (BRI_{Peak}) and (BRI_{AT}) during short, moderate and long exercise duration studies and for all data together during cycle ergometer studies. NS= no significant

Figure 6.6 demonstrates that, unlike the treadmill protocols, there is relatively poor agreement between (BRI_{Peak}) and (BRI_{AT}) during the test ramp protocols on the cycle ergometer.

Correlation between BRI and $\dot{V}_{O_2\text{ PEAK}}$ during treadmill studies.

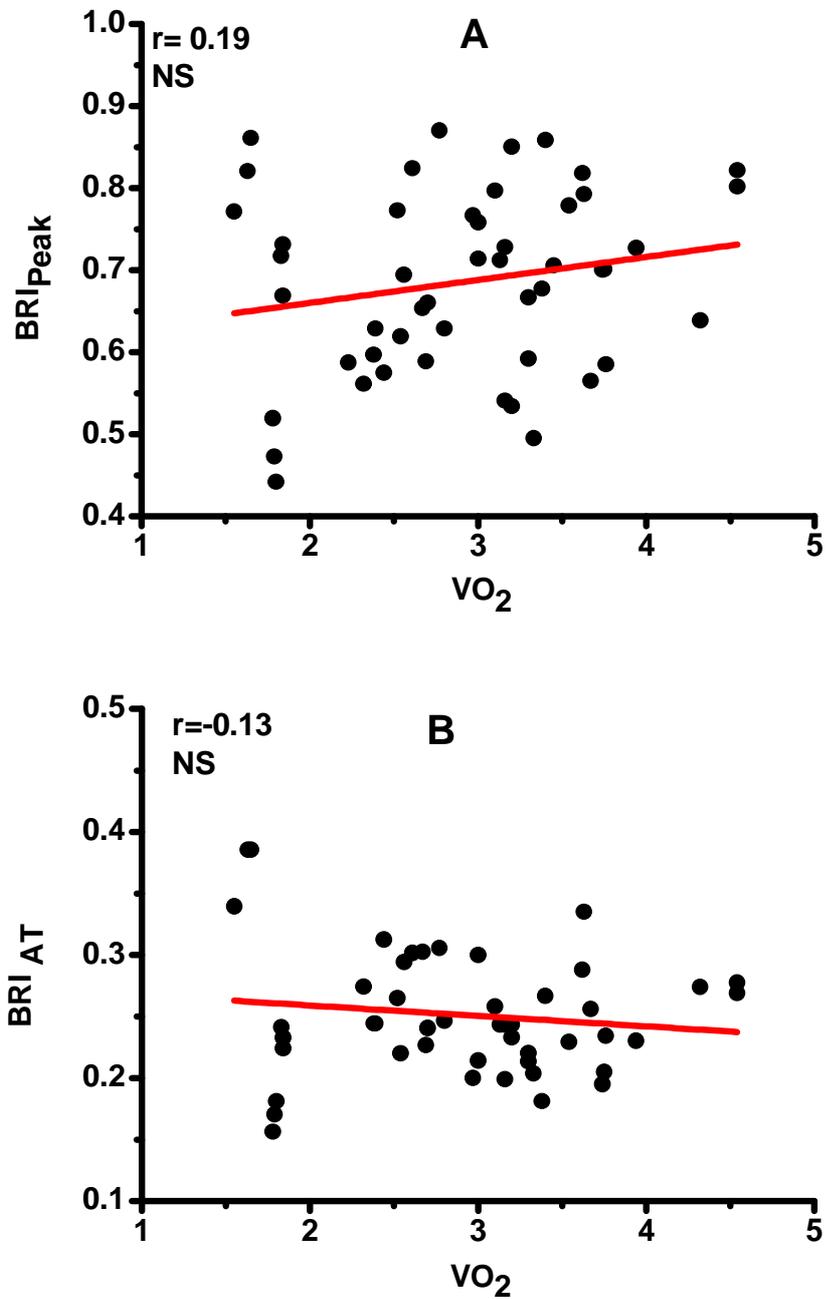


Figure 6-7 Correlation of (A) (BRI_{peak}) and (B) (BRI_{AT}) with $\dot{V}_{O_2\text{ PEAK}}$ for all data together during treadmill studies, NS = no significant correlation.

Correlation between (BRI_{Peak}) and (BRI_{AT}) with $\dot{V}_{O_2 PEAK}$ during cycle ergometer studies.

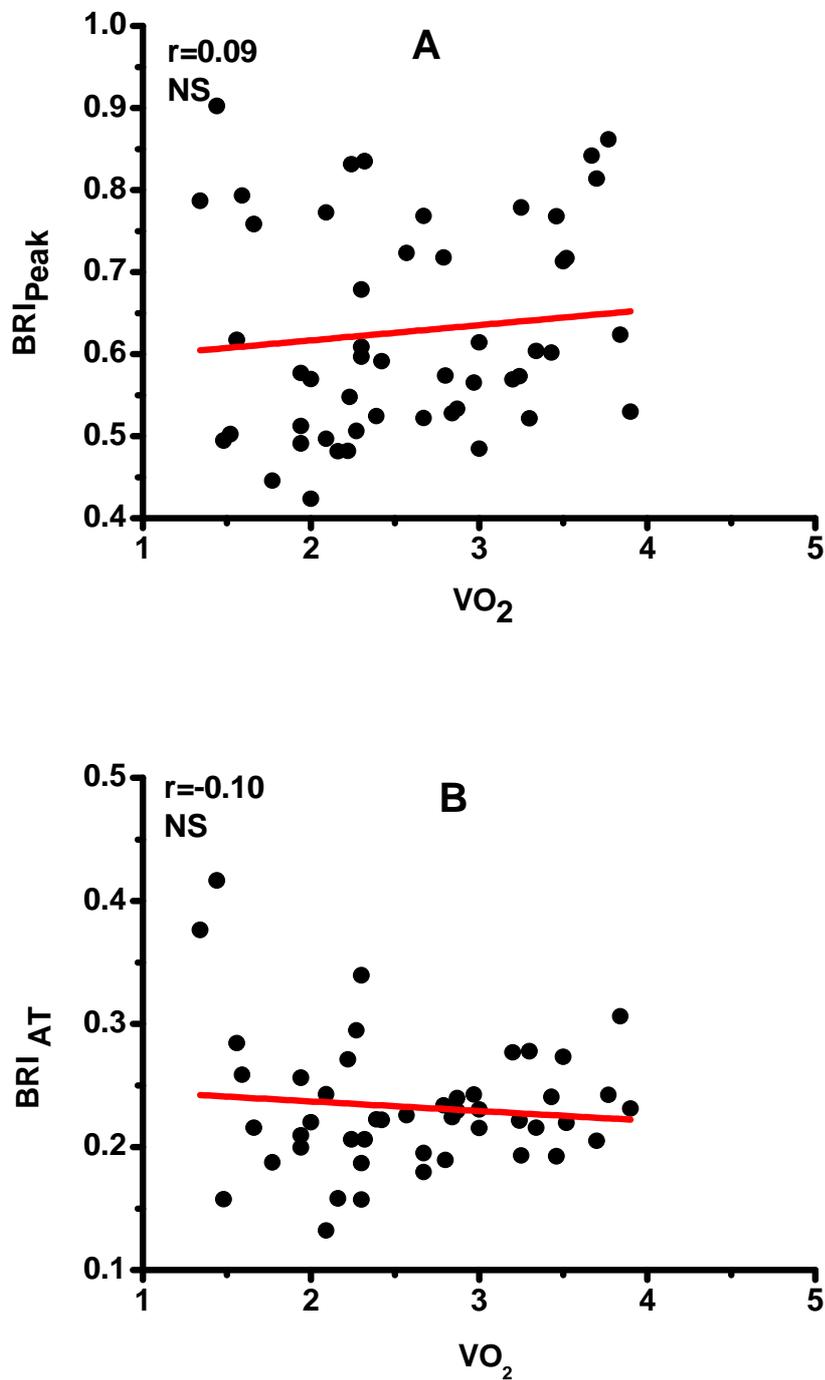


Figure 6-8 Correlation of (A) (BRI_{peak}) and (B) (BRI_{AT}) with $\dot{V}_{O_2 PEAK}$ for all data together during cycle ergometer studies, NS = no significant correlation.

Figure 6.7 and 6.8 demonstrate that there is relatively poor agreement between both measurements of breathing reserve and $\dot{V}_{O_2 \text{ PEAK}}$ during the test ramp protocols on both exercise modalities. The correlations between breathing reserve at anaerobic threshold and $\dot{V}_{O_2 \text{ PEAK}}$ were significantly weak and in the negative direction on both exercise modalities even though, male and female data were analysed together.

Relationship of breath reserve index assessment with exercise modality

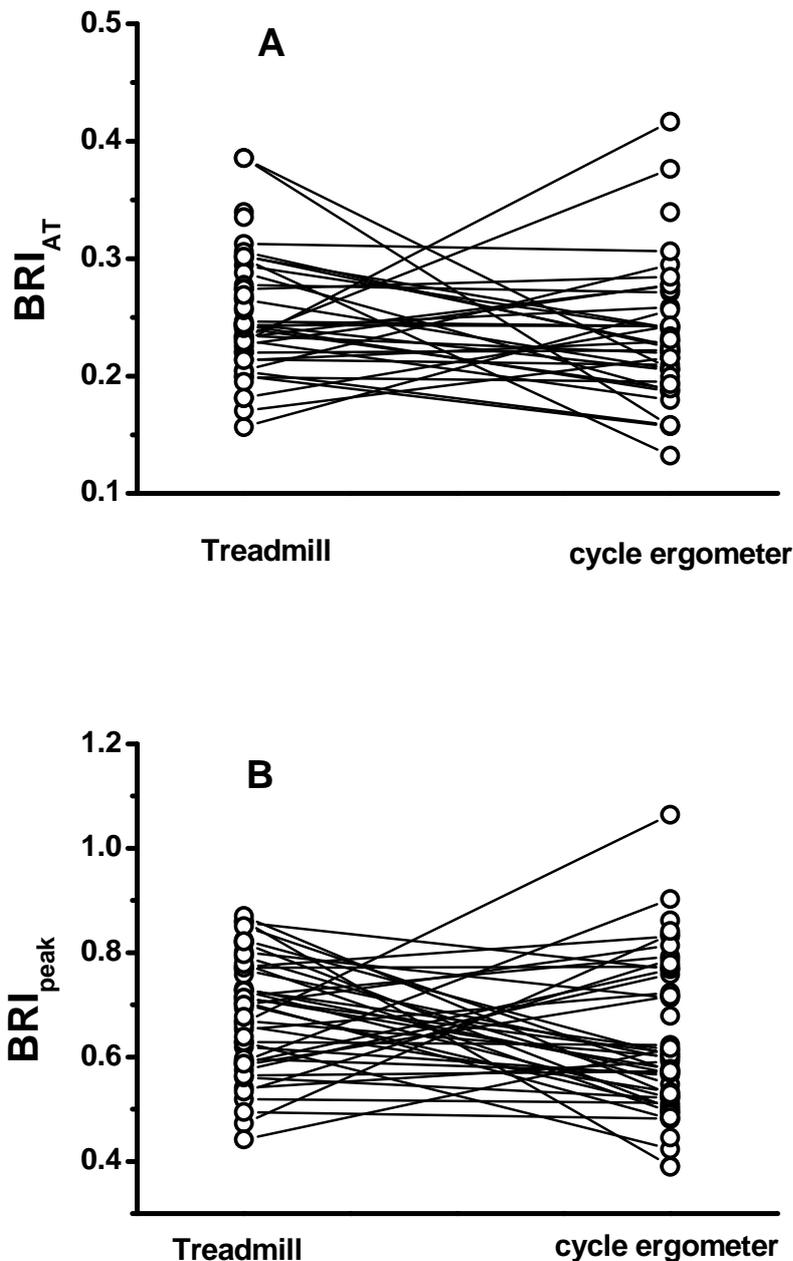


Figure 6-9 demonstrates that (A), (BRI_{AT}) is not different during treadmill and cycle ergometer studies (0.25 ± 0.05 in treadmill 0.23 ± 0.05 in cycle ergometer for all volunteers, $P=NS$), whereas (BRI_{peak}) is marginally higher during in treadmill studies but not significantly (0.69 ± 0.11 in treadmill vs 0.63 ± 0.15 in cycle ergometer for all volunteers, $P=NS$).

(Figure 6-9) demonstrates that BRI_{AT} and BRI_{peak} are independent of the type of exercise modality. In addition, both were marginally higher during treadmill exercise. That might be related to higher \dot{V}_E values during treadmill exercise. Moreover the values of BRI_{AT} was lower than BRI_{peak} in both exercise modalities.

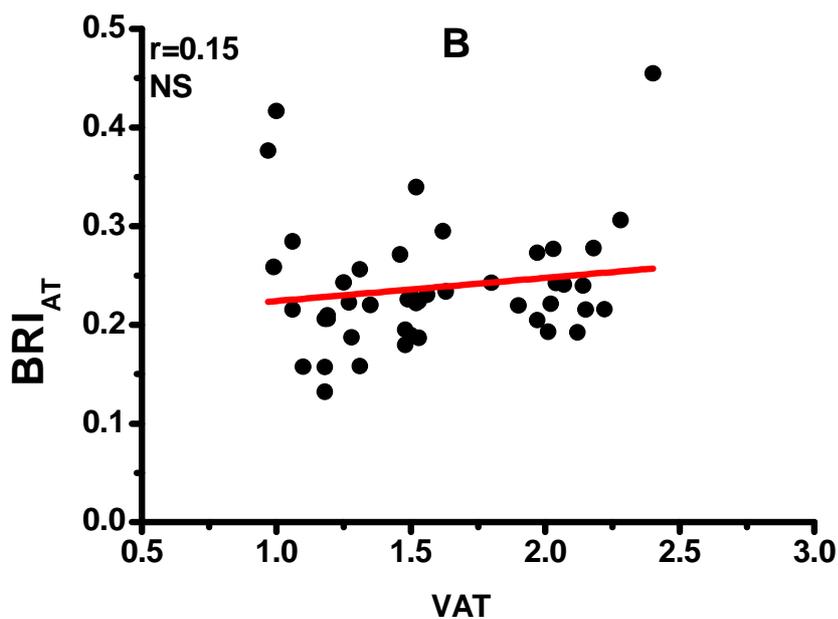
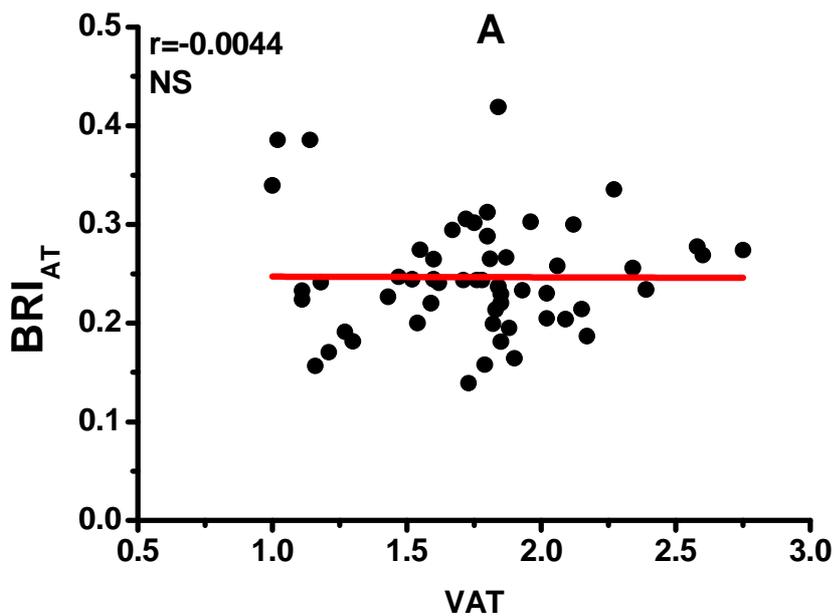


Figure 6-10 demonstrates that there is no real agreement between (BRI_{AT}) and ventilatory anaerobic threshold during the test ramp protocols on treadmill (A) and cycle ergo meter (B).

Figure 6.10 demonstrate that there is relatively poor agreement between both measurements of breathing reserve and VAT in both exercise modalities for all volunteers. Therefore, breathing reserve at anaerobic threshold cannot predict the anaerobic threshold break point.

6.7 Discussion

The aim of the current study was to directly establish whether our hypothesis that exercise duration would not influence the measurement of breathing reserve markers was correct. The data show that this is the case and that the measurement of breathing reserve index at the anaerobic threshold (BRI_{AT}) is not influenced by exercise modality. However, breathing reserve index at peak exercise is higher during treadmill exercise than in the cycle ergometer.

This difference can be attributed to the different physiological demands of cycle and treadmill exercise. Exercise on the treadmill is weight-dependent, involves a greater muscle mass for locomotion and consequently elicits a higher oxygen uptake (ATS/ACCP, 2003). The higher ventilatory requirement to support this increased aerobic demand will obviously reduce the breathing reserve available at peak exercise. While this is unsurprising as a result, the data allow some additional reflection on the potential application of breathing reserve markers in clinical practice.

Neither of the markers of breathing reserve correlated well with peak oxygen uptake, even when higher proportions of the breathing reserve were used during treadmill exercise (data not shown) and suggest that, like the oxygen uptake efficiency slope in chapter 5, this measurement may be a useful clinical tool that may stratify functional capacity but not predict aerobic performance. This may be related to the relative contribution that gross pulmonary function contributes to the determinants of peak oxygen uptake (where it is generally considered that cardiac output is the major determinant) (Astrand & Rodahl, 1986;ATS/ACCP, 2003).

It is likely that this lack of specificity will be more apparent in patient populations with pulmonary disorders. In such patients, the pulmonary limitations may cause the cessation of physical activity (in the real world situation) or exercise (in cardiopulmonary exercise testing) before the limits on the cardiovascular system are challenged. Since the pulmonary limitations provide a lower order determinant of oxygen uptake, the relative difference in peak oxygen uptake will be reduced when this system fails (*i.e.* the breathing reserve is used up before the wide distribution in oxygen uptake can be observed

and therefore specificity is lost). This limitation is supported by the observation in previous studies where, in contrast to the present study, (BRI_{AT}) correlates with (BRI_{Peak}) in COPD and respiratory limited cardiac patients (in this previous study $r=0.85$, $p<0.0001$) (Medoff *et al.*, 1998) and Cystic Fibrosis patients (in this previous study $r=0.89$, $p<0.01$) (Sexauer *et al.*, 2003).

In both the previous studies, the strong positive correlations reflect a high (BRI_{AT}) in the patient populations (around 0.6 in both). This relatively high (BRI_{AT}) is measured because the patients exercise is limited by their pulmonary system which fails soon after the anaerobic threshold. This means that (BRI_{Peak}) follows closely on from the anaerobic threshold, providing a good correlation between the two measurements that is less apparent when exercise is not limited by the respiratory system. This observation further emphasises the likelihood of dissociation between the breathing reserve and oxygen uptake and, importantly, lower values for the (BRI_{AT}) measured in the present study may have identified another use for this marker of functional capacity.

Ventilation is obviously driven by carbon dioxide concentrations in the blood and ventilation increases disproportionately with oxygen uptake at exercise intensities above the lactate threshold (Davis *et al.*, 1976; Aunola & Rusko, 1986). This could mean that (BRI_{AT}) would be a useful non-invasive predictor of ventilatory anaerobic threshold. The data presented here show that (BRI_{AT}) was similar during short, moderate and long exercise duration. Thus, (BRI_{AT}) is a consistent and reproducible measurement which is independent of exercise duration and/or modality. However, (BRI_{AT}) does not correlate with the ventilatory derived anaerobic threshold. This observation suggests that (BRI_{AT}) is not a useful marker of anaerobic capacity in patient populations.

These data confirm that breath reserve is useful marker of functional capacity in patients with respiratory limitations but that it is, perhaps, less appropriate for healthy populations. The measurement is not influenced by exercise duration or exercise modality, despite a higher ventilation in treadmill exercise which emphasises a lack of specificity.

These data, and those detailed in the previous experimental chapters demonstrate that the current American College of Sports Medicine guidelines are

too prescriptive for clinical exercise testing. Accurate measurement of clinically relevant markers of functional capacity can be achieved during short and long duration exercise testing. The limit to the accuracy of the measurement is much more related to the administration of the exercise protocol and the use of appropriate analysis techniques to determine an accurate measurement of the chosen marker. Avoiding pre-exercise hyperpnoea (by delaying the start of the protocol until a normal resting ventilation and respiratory exchange ratio is observed) and including a phase of unloaded exercise is much more important to the outcome of the test than exercise duration. The importance of such procedures should be included in subsequent guidelines.

CHAPTER SEVEN

General Discussion

7 General Discussion

The physiological response to a cardiopulmonary exercise test (CPET) is influenced by the nature of the exercise protocol and the exercise modality employed (Hansen *et al.*, 1988; Hermansen & Saltin, 1969; Workman & Armstrong, 1963). In relation to exercise modality, the most common exercise modalities used in clinical exercise testing laboratories are treadmills and cycle ergometers. As the biomechanics of walking (weight dependent) and cycling (weight independent) are completely different it was important to investigate them as separate entities. Furthermore, the recent shift in clinical exercise testing laboratories from step exercise protocols (with 3 minute stages) to ramp increment protocols has focused attention on the appropriateness of test protocols employed. It is important to ensure the accuracy and comfort for patients during the determination of maximal and sub-maximal testing (Myers & Bellin, 2000; Myers *et al.*, 2001).

The main aim of this thesis was to assess the validity of the current ACSM guidelines for clinical exercise testing. The desired outcome was to obtain protocols that allowed participants to obtain the peak exercise response within exercise durations that conformed to the guidelines (between 8-12 minutes) or that were shorter or longer. The reality was that this was difficult to achieve in patients on both exercise modalities due to the heterogeneous study population.

The ACSM guidelines are based on a single study (Buchfuhrer *et al.*, 1983) where the best five subject responses from different protocols were used to establish the appropriate exercise durations. However, in clinical practice there are standardised protocols employed to use exercise duration as a marker of exercise performance (so that each hospital laboratory will recognise the exercise duration as workload intensity).

This approach means that it is only in research laboratories where protocols are refined to try and obtain peak exercise within the 8-12 minute time scale in all patients. In the clinical environment, many patients do reach their maximum within this period but a significant proportion exercise for shorter or longer durations and consequently the validity of the test is called into question. The variability in exercise duration reflects the broad spectrum of physiological

limitations observed in clinical laboratories during the progression of cardiopulmonary or other diseases that limit exercise capacity.

Thus, the data presented in this thesis is more relevant to clinical exercise testing than the Buchfuhrer *et al* study (Buchfuhrer *et al.*, 1983). The findings that exercise durations shorter or longer than the current ACSM guidelines do not significantly influence the measurement of peak cardiopulmonary exercise variables have important implications for the validity of clinical exercise testing in it's current format.

These data support the previous assertions that short exercise durations can, in clinical practice, provide useful maximal data for frail patients (Redwood *et al.*, 1971) and that unnecessary prolongation of exercise duration may have a negative impact on patients (Northridge *et al.*, 1990). This suggests that the current guidelines are too prescriptive in relation to exercise test duration. However, the guidelines may have had some inadvertent value because the current guidelines do not address issues related to sub-maximal exercise performance analysis (which may be more appropriate in some clinical populations).

Further analysis of the data available from the studies conducted in this thesis has, for the first time, examined the influence of exercise duration on sub-maximal markers of exercise performance. The concern would be that although short exercise durations do not limit the validity of maximal data; the limited data volume available for sub-maximal analysis might be a problem. This is a concern because much of this analysis is performed via linear regression where the quality of the data fitting procedures can be compromised by low data volume. The data presented here, however, show that despite the expected reduction in data quality there is no significant influence of changing exercise duration on the validity of the commonly used sub-maximal markers in clinical practice (*i.e.* ventilatory threshold, breathing reserve and oxygen uptake efficiency).

There are some limitations in the data presented in this thesis to support the aim to assess the validity of the current ACSM guidelines for clinical exercise testing. The data does provide a greater age range, a more diverse range in exercise capacity and includes females in the analysis; all of which improves on

the study that underpins the current guidelines (Buchfuhrer *et al.*, 1983). However, the lack of clinical populations in the analysis still limits the relevance of this data to support good evidence based clinical guidelines.

At the outset, the intention was to develop a robust exercise protocol that could use the same workload increments and alter the 'step duration' to provide a response where only exercise duration changed. The pilot studies demonstrated that this was a bigger task than originally envisaged and the additional time taken to complete these tests meant that additional studies were unachievable within the time available. In addition, even when a final protocol was chosen it became apparent that not enough participants could achieve exercise durations that precisely conformed to the ACSM guidelines, were shorter and/or longer. This problem meant that some participants were only available for tests on one exercise modality and reduced the power of our studies for statistical analysis despite a very significant level of recruitment in total.

Additionally, a major issue that limits the validity of sub-maximal markers in CPET is pre-exercise hyperpnoea. A significant attempt to prevent this issue was made by using significant rest and unloaded exercise breath-by-breath measurements during the test protocols. These periods aimed to ensure that the participants' breathing frequency and ventilation were in an acceptable range before the start of the exercise protocol. However, the vast majority of the study participants were unaccustomed to exercise physiology testing and some patients were excluded because of significant pre-exercise hyperpnoea. This again reduced statistical power despite a very significant level of total recruitment.

Furthermore, measuring the maximal voluntary ventilation (MVV) is problematic (Kor *et al.*, 2004). The technique requires significant co-operation from participants and valid MVV measurements could not be obtained in all participants. For this reason, many previous studies estimate maximum voluntary ventilation from the forced expiratory volume in 1 second (FEV₁) (Gandevia & Hugh-Jones, 1957; Miller *et al.*, 1959; Kor *et al.*, 2004) instead of using direct measurement (Dillard *et al.*, 1993). The relevance of this data to other studies would have been improved by including estimated MVV in the analysis.

Despite these limitations, however, the data presented here do add to our current understanding of the factors limiting the validity of CPET data. They provide additional information that highlights limitations to the current guidelines for clinical exercise testing and raise issues that provide a focus for future research. For example, while it is important to take such validation into the clinically relevant populations it is apparent that identifying robust protocols to ensure a consistent response in exercise duration is difficult in heterogeneous groups. As exercise testing is not without risk in clinical populations it will be prudent to undertake pilot experiments to determine appropriate exercise protocols in the relevant exercise modalities. Much of this pilot work could be achieved, without risk to patients, by modeling factors that limit exercise performance in healthy participants matched for age and with relatively low exercise capacity (unfit rather than unhealthy). Such interventions could include artificially increasing the physiological dead space to limit pulmonary function, reducing autonomic function with beta-blockers to limit cardiovascular function or reducing inspired oxygen content to limit both pulmonary and cardiovascular reserve. These relatively safe interventions would allow targeted protocols to be applied in clinical populations, reducing the risk to the patient by lowering the number of tests required to validate maximal and sub-maximal markers of exercise performance at different exercise durations.

These data would allow the ACSM guidelines to be modified and provide a better rationale for evidence based practice in clinical exercise testing. However, even without these additional experiments it is evident from the data presented here that the current ACSM guidelines are too prescriptive for clinical exercise testing. Valid measurement of clinically relevant markers of functional capacity can be achieved during short and long duration exercise testing. The limit to the accuracy of the measurement appears to be much more related to the technical application of the exercise protocol and the use of appropriate analysis techniques. Avoiding pre-exercise hyperpnoea (by delaying the start of the protocol until a normal resting ventilation and respiratory exchange ratio is observed) and including a phase of unloaded exercise is, at present, much more important to the outcome of the test than exercise duration. The importance of such procedures should be included in subsequent guidelines.

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8 Appendices

Appendix 1

Consent Form



**UNIVERSITY
of
GLASGOW**

CONSENT FORM

The impact of exercise duration on sub-maximal ‘markers’ of exercise performance

SUBJECT NAME.....DATE OF BIRTH.....

To be completed by the Subject

Please Tick

- | | Yes | No |
|--|--------------------------|--------------------------|
| Have you read the Information Sheet Version (enter Version number/date)? | <input type="checkbox"/> | <input type="checkbox"/> |
| Have you had an opportunity to ask questions and discuss this study? | <input type="checkbox"/> | <input type="checkbox"/> |
| Have you received satisfactory answers to all your questions? | <input type="checkbox"/> | <input type="checkbox"/> |
| Have you received enough information about the study? | <input type="checkbox"/> | <input type="checkbox"/> |

Who have you spoken to?
Prof/Dr/Mr/Ms_____

- | | | |
|---|--------------------------|--------------------------|
| Do you understand that you are free to withdraw from the study at any time and without having to give a reason? | <input type="checkbox"/> | <input type="checkbox"/> |
| Do you agree to take part in this study? | <input type="checkbox"/> | <input type="checkbox"/> |

Signed..... Date.....

.....

Name in Block

Letters.....

Signature of _____ Date.....

Witness.....

Name in Block

Letters.....

Appendix 2

Information Sheet

INFORMATION SHEET

TITLE OF INVESTIGATION: The impact of exercise duration on sub-maximal 'markers' of exercise performance.

You are invited to take part in a research study looking at the impact of exercise duration on sub-maximal measurements of exercise performance. In order to help you to understand what the investigation is about, please read the following information carefully. If there are any points that need further explanation, please ask me. It is important that you understand what you are volunteering to do and are completely happy with all the information before you sign this form.

What is the purpose of the study? The purpose of this study is to gather information about how the duration of exercise will influence the measurements being made during medical testing. Exercise is commonly used in hospitals as part of the procedures used in the diagnosis of many medical conditions. There are guidelines on how to conduct such tests that focus on how to make sure that the patient reaches maximum effort, however, this is not possible in all patient groups and we hope to provide information that will allow these guidelines to be modified to provide reliable data without patients making maximum effort.

Why have I been chosen? You have been selected as a possible participant in this investigation because you are healthy man or woman within the specified age range and are able to participate in treadmill or cycle exercise.

Do I have to take part? It is up to you to decide whether or not to take part. If you decide to take part, you will be given this information sheet to keep and you will be asked to sign a consent form. If you decide to take part, you are still free to withdraw at any time and without giving a reason.

What will happen to me if I take part? You will be asked to come along to the University of Glasgow to be introduced to the staff and receive further information about the tests being performed in this study. At this time you will complete a health questionnaire or be screened by a doctor (if appropriate) to ensure that you are able to take part in the study. Once this initial screening is complete, you will be asked to complete three maximal exercise tests on a treadmill or a cycle. These tests should last no more than 30 minutes and a single visit will last less than one hour (the 3 visits should be completed within a 3 week period).

On completion of the 3 exercise tests, we will ask if you would be willing to undertake another 3 tests (on a cycle if you completed the treadmill tests or *vice versa*). If you agree to complete further tests, you will not be asked to attend within one month of the first batch of tests (to prevent the inconvenience of you attending the exercise laboratory on too many occasions within a short period).

Is the research part of a student's coursework? Yes. The research will form a major part of the submission for a post-graduate student's PhD.

What are the side effects of taking part? There are no foreseen side effects to taking part in this research.

What are the possible disadvantages and risks of taking part? Little risk is envisaged. While exercise carries a small risk of injury the screening process should ensure that you are fit to complete the tests. Staff are trained in first aid and have emergency procedures in place should any accident occur. Your anonymity will be maintained in any reports that emerge from this work. No disadvantages are foreseen.

What are the possible benefits of taking part? The benefits to be gained from the study could be substantial in regard to understanding the best way to conduct clinical exercise testing. More robust and meaningful data from clinical exercise testing will help in developing a diagnosis and/or prognosis for patients attending hospital clinics. The gains are therefore far greater than the negligible risks involved for the participants.

What if something goes wrong? The potential risks involved in your participation in the study have been outlined above. If you are harmed by taking part in this research project, there are no special compensation arrangements. If you are harmed due to someone's negligence, then you may have grounds for a legal action but you may have to pay for it.

Will my taking part in this study be kept confidential? All information which is collected about you during the course of the research will be kept strictly confidential.

Is the research funded? No.

What will happen to the results of the research study? The results of the study will be published (both in a PhD thesis and in a peer reviewed medical journal). It will not be possible to identify you from the data presented in any publication. You can be sent a copy of the full report if you wish.

If you are worried about any of the issues involved in the above procedures, you should contact Dr Niall MacFarlane (via e-mail at N.MacFarlane@bio.gla.ac.uk or phone at 0141-330-5965).

Appendix 3

Physical activity questionnaire

UNIVERSITY OF GLASGOW
FACULTY OF BIOMEDICAL AND LIFE SCIENCES
SUBJECT QUESTIONNAIRE FOR HIGH INTENSITY EXERCISE TESTING

If you feel unwell on the day of a proposed test, or have been feeling poorly within the last two weeks, you are excluded from taking part in an exercise test. The considerations that follow apply to people who have been feeling well for the preceding two weeks.

NAME

SEX: M/F AGE: (yr) HEIGHT: (m) WEIGHT: (kg)

Details of last medical examination (where appropriate):

Date: Location:
 (day/mo/yr)

Exercise lifestyle:

What kind(s) of exercise do you regularly do (20 min or more per session), and how often?
 (Please circle the number of times per average week):

Walking	1	2	3	4	5
Running	1	2	3	4	5
Cycling	1	2	3	4	5
Swimming	1	2	3	4	5
Skiing	1	2	3	4	5
Rowing	1	2	3	4	5
Gymnastics	1	2	3	4	5
Martial Arts					
Tune Up	1	2	3	4	5
Popmobility	1	2	3	4	5
Sweat Session	1	2	3	4	5
Weight Training	1	2	3	4	5
Field Athletics	1	2	3	4	5
Racket Sports	1	2	3	4	5
Rugby/soccer/hockey	1	2	3	4	5
Others*	1	2	3	4	5

*(Please specify)

How long have you been exercising at least twice/week for at least 20 min/session?
 years

Smoking: (Please tick one) Never smoked Not for >
 6 months
 Smoke < 10 per day Smoke > 10 per day

Illnesses: Have you ever had any of the following? (Please circle NO or YES)

Anaemia	NO/YES	Asthma	NO/YES
Diabetes	NO/YES	Epilepsy	NO/YES
Heart Disease	NO/YES	High Blood Pressure	NO/YES
Other*	NO/YES		

*(Please specify)

Symptoms:

Have you ever had any of the following symptoms to a significant degree *at rest or during exercise*? That is, have you had to consult a physician relating to any of the following?

	<i>Rest</i>	<i>Exercise</i>
Breathlessness	NO/YES	NO/YES
Chest Pain	NO/YES	NO/YES
Dizzy Fits/Fainting	NO/YES	NO/YES
Heart Murmurs	NO/YES	NO/YES
Palpitations	NO/YES	NO/YES
Tightness in chest, jaw or arm	NO/YES	NO/YES
Other*	NO/YES	

*(Please specify)

Muscle or joint injury:

Do you have/or have had any muscle or joint injury which could affect your safety in performing exercise (*e.g. cycling or running*), strength testing or strength training? NO/YES*

*(Please specify)

Medication:

Are you currently taking any medication? NO/YES*

*(Please specify)

Family History of Sudden Death:

Is there a history of sudden death in people under 40 years in your family? NO/YES*

Can you think of any other reason why you should not take part in our tests? NO/YES*

(Please specify)

The following exclusion and inclusion criteria will apply to this study:

Exclusion Criteria

If you have any of the following, you will be excluded from the study:

- Asthma, diabetes, epilepsy, heart disease, a family history of sudden death at a young age, fainting bouts, high blood pressure, anaemia and muscle or joint injury.
- If you are taking any medication that may adversely affect your performance or health in this study, you will not be allowed to take part in the study.
- If you take recreational drugs, you will not be allowed to take part in the study.
- If you have ingested alcoholic drinks in the previous 48 hours, you will not be allowed to take part in the study.

Inclusion Criteria

- Male or female subject aged at least 18 years.
- In good health at the time of testing.

SignatureDate

Body Weight and Blood Pressure:

Body Weight: Height:

BP (Resting)

Screened by: Date:

Appendix 4

Approved Ethics Proposal

**Ethics Committee for Non Clinical Research
Involving Human Subjects, Material or Data**

Application Form for Ethical Approval

NOTES:

A submission to this Committee does not automatically result in approval. Investigators must wait for written approval before commencing data collection. Disciplinary measures will be taken if work commences without ethical approval being in place. The matter will be referred to the Dean for appropriate action.

THIS APPLICATION FORM SHOULD BE TYPED, NOT HAND WRITTEN.

ALL QUESTIONS MUST BE ANSWERED. "NOT APPLICABLE" IS A SATISFACTORY ANSWER WHERE APPROPRIATE.

Project Title: The impact of exercise duration on sub-maximal 'markers' of exercise performance

Is this project from a commercial source, or funded by a research grant of any kind? No

If yes,

a) Has it been referred to Research & Enterprise?
Has it been allocated a project Number?

b) Give details and ensure that this is stated on the Informed Consent form.

Date of submission: 7/5/09

Name of all person(s) submitting research proposal

- 1) Dr Niall Gordon MacFarlane
- 2) Mr Abdulrahman Al Howikan

Position(s) held

- 1) Senior Lecturer
- 2) Post-graduate student

Division

Integrative and Systems Biology

Address for correspondence relating to this submission
Room 240A, West Medical Building, University of Glasgow G12 8QQ

Name of Principal Researcher (if different from above e.g., Student's Supervisor):

Position held :

1. Describe the purposes of the research proposed.

Assessing exercise tolerance is a common technique applied by many physicians for diagnostic and prognostic purposes in a range of clinical conditions¹. Common testing procedures will, in general, conform to the American College of Sports Medicine guidelines for exercise testing and aim for the patient or experimental participant to reach their maximum exercise capacity within 8-12 minutes (and not to exceed 15 minutes)².

The rationale for this exercise duration is that it facilitates an accurate measurement of the peak oxygen consumption (peak VO_2). This criterion originated from the findings of a single study by Buchfuhrer *et al* (1983) where five young, healthy, male subjects performed five treadmill and three cycle ergometer tests and highest peak VO_2 s were recorded with a work rate increment that brought the subjects to the limit of their tolerance in 10 ± 2 minutes³. In this study, however, longer test durations did not result in any difference in the peak VO_2 measured but were discounted because the authors saw no benefit in prolonging the test duration.

We believe, however, that in older and infirm populations there are substantial benefits from using sub-maximal markers of aerobic performance and that longer duration incremental exercise tests may have more practical value. To this end, we propose to study the effects of exercise duration on peak and sub-maximal markers of exercise performance to challenge current dogma.

The research proposed here will study the effects of exercise duration on peak VO_2 , lactate threshold, ventilatory threshold, breathing reserve at the lactate threshold and the oxygen uptake efficiency slope in male and female populations over a wide age range. Later studies will simulate the pathophysiology of common medical conditions in healthy volunteers before validation in clinical populations to provide significant data to support proposals for amendments to the current guidelines.

References

1. Karlman Wasserman, James E. Hansen, Darryl Y. Sue, William W. Stringer and Brian J. Whipp. Principles of exercise testing and interpretation - 4th Edition published by Lippincott, Williams and Wilkins (ISBN: 9780781748766).
2. American College of Sports Medicine. ACSM guidelines for exercise testing and prescription. - 8th Edition (ISBN: 0781769027).
3. Buchfuhrer MJ, Hansen JE, Robinson TE et al. Optimizing the exercise protocol for cardiopulmonary assessment. J Appl Physiol 1983; 55: 1558-64.

2. Please give a summary of the design and methodology of the project. Please also include in this section details of the proposed sample size, giving indications of the calculations used to determine the required sample size, including any assumptions you may have made. (If in doubt, please obtain statistical advice).

The study has a simple design, to perform cardiopulmonary exercise testing using protocols that last less than the ACSM guidelines (reaching maximum exercise capacity between 8 and 12 minutes), that conform to the ACSM guidelines and that last longer than the ACSM guidelines. This would require participants to perform exercise on three occasions, no longer than one week apart and in random order using 3 standardized exercise protocols. While exercise modalities are different throughout the world, the main approaches use either a cycle ergometer or a treadmill. The biomechanics of walking and cycling are different and may influence the physiological measurements being proposed. It is our intention, therefore, to duplicate these tests using a cycle ergometer and a treadmill (so that the participants would need to visit the lab six times for data collection purposes).

Treadmill studies

The three protocols proposed for data collection use a modification of the ramp Bruce protocol¹. The normal stage duration for this protocol is 15s and will be reduced to 10s to reach maximum in less than the ACSM guidelines or increased to 20s to reach maximum in more than the ACSM guidelines. During exercise the expired gas will be collected using standard techniques and analyzed for the expired concentrations of O₂ and CO₂ on a breath-by-breath basis (using the Oxycon gas analysis system from Viasys). This analysis will allow us to determine the effects of exercise duration on peak oxygen consumption (like Buchfuhrer *et al*, 1983) but more importantly to assess the affect on sub-maximal markers of exercise performance that may be more relevant in clinical populations. This study will allow us to measure the effects of exercise duration on oxygen uptake efficiency slope, breathing reserve at the lactate threshold and the ventilatory threshold (all commonly used non-invasive markers of exercise performance). As stated previously, the protocols will be applied in random order, on three separate occasions no more than one week apart.

Cycle ergometer studies

There is a common split in exercise modalities used in clinical practice where the UK and US tend to use treadmill exercise and mainland Europe tends to use cycle exercise. We propose to duplicate the treadmill study using cycle ergometer exercise. The participants will be asked to attend a separate set of exercise sessions (not within one month of the treadmill tests to limit their inconvenience). The participants will be asked to perform three cycle protocols, a ramp protocol of 20W/min to reach their maximum in less than the ACSM guidelines, a ramp protocol of 15W/min to conform to the ACSM guidelines and a ramp protocol of 10W/min to reach their maximum in more than the ACSM guidelines. The expired gases will be analyzed in the same way as described for the treadmill protocols.

Age/Gender of groups to be investigated

It is well known that the age and gender of participants does not influence the slope of the work rate to oxygen consumption relationship. However, the starting position of this relationship is under the influence of age and gender and could effect the exercise duration. For this reason, we propose to analyze the effects of exercise duration on the physiological parameters being measured in separate cohorts of age and gender. Thus we will recruit both male and females healthy volunteers in the following age ranges (18-29, 30-44 and 45-60). Standard power calculations suggest that 12 subjects in each gender and age category will suffice for comparing the effects of exercise duration in the exercise protocols using Bland-Altman plots.

1. Kaminsky LA and Whaley MH (1998) Evaluation of a New Standardized Ramp Protocol: The BSU/Bruce Ramp Protocol *Journal of Cardiopulmonary Rehabilitation* 18(6): 438-444.

3. Please give a detailed account of the method of recruitment of volunteers.

Study participants will be recruited from the University staff population and the general public using advertisements on University websites and the local media. All volunteers will be sent information packs (including information on informed consent before attending for a health screening and familiarization visit).

4. Describe the research procedures as they affect the research subject and any other parties involved.

The procedures involved in this project are standard techniques for assessing health status and exercise performance in the general population. There is a low risk of injury in taking part in maximal exercise testing that increases with age. The younger cohort (18-29 years) will be screened using a general health questionnaire that will highlight any particular risks in performing exercise (any participant at risk will be excluded from the study). The 'middle age group' of participants (30-44 years) will be screened by a medical practitioner, if over 35 years, or by a general health questionnaire and any participant at risk will be excluded from the study. Participants from the 'older' age range will be screened by a medical practitioner prior to exercise and will be monitored by a physician during the exercise tests.

The participants may have to attend the lab on at least 7 occasions during this study (1 screening and familiarization visit and six data collection visits). This is a significant commitment and as such we will not ask them to attend for treadmill or cycle exercise bouts in one block. It would also be possible for participants to attend for only cycle or treadmill exercise (and, as usual, they could withdraw from the study at anytime without giving a reason).

There is no invasive sampling to be performed as part of this study and the risks to investigators are minimal.

5. What in your opinion are the ethical considerations involved in this proposal? (You may wish for example to comment on issues to do with consent, confidentiality, risk to subjects, etc.)

The participants will all be healthy adults and will go through the process of informed consent. No participants will be in a dependant relationship with the investigators and all data will be held anonymously. There is a small risk of injury in performing maximal exercise but with appropriate screening any adverse event is highly unlikely.

6. Outline the reasons which lead you to be satisfied that the possible benefits to be gained from the project justify any risks or discomforts involved.

The practice of cardiopulmonary exercise testing is common throughout the world and has a significant impact on resources for health care in the developed world. It is important to be sure that the guidelines for exercise testing protocols are suitable for practical application in clinical testing. This study will provide evidence to support proposals for amending the current guidelines and may provide appropriate protocols for clinical exercise testing.

7. Who are the investigators (including assistants) who will conduct the research and what are their qualifications and experience? Is it necessary for the investigators to have clearance from Disclosure (Scotland) and if so, has this been completed?

No disclosure is necessary but all investigators will make basic disclosure applications..

Mr Abdurahman Alhowikan will be responsible for conducting these studies as part of the research towards his PhD. He has a background of clinical exercise testing and over 5 years experience in conducting exercise tests. Mr John Wilson will provide technical support for these studies and has over 25 years experience in exercise testing. Dr Niall MacFarlane will supervise this project and has more than 25 years experience in clinical exercise testing.

8. Are arrangements for the provision of clinical facilities to handle emergencies necessary? If so, briefly describe the arrangements made.

The technical and academic supervisors named in this application have basic life support and first aid qualifications. Any significant medical emergency will be dealt with using standard University procedures. Where the 'older' participants in this study are involved the tests will be supervised by a clinician to supplement standard protocols.

9. In cases where subjects will be identified from information held by another party (for example, a doctor or hospital) describe the arrangements you intend to make to gain access to this information including, where appropriate, which Multi Centre Research Ethics Committee or Local Research Ethics Committee will be applied to.

N/A

10. Specify whether subjects will include students or others in a dependent relationship and where possible avoid recruiting students who might feel to be or be construed to be under an obligation to volunteer for a project. This is most likely to be where a student is enrolled on a course where the investigator is a teacher. In these circumstances the recruitment could be carried out by one of the other investigators or a suitably qualified third party.

Current students will not be excluded from recruitment, but given the age ranges being recruited it is unlikely that students in a dependant relationship will be recruited and there will be no pressure to have to recruit from this population.

11. Specify whether the research will include children or people with mental illness, disability or handicap. If so, please explain the necessity of involving these individuals as research subjects.

No

12. Will payment or any other incentive, such as a gift or free services, be made to any research subject? If so, please specify and state the level of payment to be made and/or the source of the funds/gift/free service to be used. Please explain the justification for offering payment or other incentive.

No

13. Please give details of how consent is to be obtained. A copy of the proposed consent form, along with a separate information sheet, written in simple, non-technical language MUST ACCOMPANY THIS PROPOSAL FORM.

Informed consent will be obtained at the first screening, familiarization, visit. An information sheet will be sent to participants before their attendance at this visit to allow them to make a decision on their participation before that visit.

14. Comment on any cultural, social or gender-based characteristics of the subject which have affected the design of the project or which may affect its conduct.

N/A

15. Please state who will have access to the data and what measures which will be adopted to maintain the confidentiality of the research subject and to comply with data protection requirements e.g. will the data be anonymised?

The named investigators will have access to the data. All data will be anonymised and copies will be held on secure databases with hard copies retained in a locked filing cabinet.

16. Will the intended group of research subjects, to your knowledge, be involved in other research? If so, please justify.

No

17. Date on which the project will begin 1st July 2009 and end 31st December 2010.

18. Please state location(s) where the project will be carried out.

West Medical Building, Glasgow University.

19. Please state briefly any precautions being taken to protect the health and safety of researchers and others associated with the project (as distinct from the research subjects) e.g. where blood samples are being taken

Standard operating procedures for the Institute of Diet, Exercise and Lifestyle exercise laboratories will be adhered to at all times. No invasive measurements are being undertaken and consequently the risks to the investigators are low.

Signed _____ Date _____
(Proposer of research)

Where the proposal is from a student, the Supervisor is asked to certify the accuracy of the above account.

Signed _____ Date _____
(Supervisor of student)

Email the completed form to: S.Morrison@bio.gla.ac.uk

And send the signed hard copy to:

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