MEASURING ANAEROBIC PERFORMANCE IN CHILDREN USING ACCUMULATED OXYGEN DEFICIT

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DEDICATION

This thesis is dedicated to the memory of my brother Andrew, whose passing awakened my mind and opened my heart.

Until we meet again in the light.....

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ASSESSING THE ANAEROBIC PERFORMANCE OF CHILDREN USING ACCUMULATED OXYGEN DEFICIT

Abstract

The thesis investigated a method of determining accumulated oxygen deficit (AOD) in children and quantified the anaerobic performance of children in a variety of ways. The first of the studies investigated the developmental profile of anaerobic performance using AOD in three groups of male children classified as pre-pubertal, pubertal or post-pubertal. AOD ($\sum$L) of the post-pubertal group (3.97 ± 0.58) was greater that that of the pubertal (2.88 ± 0.42) group and the pre-pubertal (1.38 ± 0.21) group ($p<0.05$). The results from the first study suggest that AOD is sensitive enough to detect differences in the anaerobic performances of the different groups, but the methodology used to determine AOD proved to be time-consuming. In response to the time-consuming nature of the methodology used to determine AOD in Study One, the aims of Study Two were twofold. The first aim was to investigate potential differences in the oxygen uptake when discrete and continuous incremental submaximal protocols were used to determine the relationship between submaximal oxygen uptake and exercise intensity. The second aim was to investigate the repeatability of oxygen uptake during continuous incremental submaximal exercise. Two groups of pre-pubertal males participated in the testing sessions. The results of Study Two found no differences in the submaximal oxygen uptake at treadmill speeds of 6 km.hr^{-1}, 7 km.hr^{-1}, 8 km.hr^{-1} and 8.5 km. hr^{-1} when discrete and continuous incremental submaximal exercise protocols were compared. There were no differences in the linear regression equations calculated using the relationship between submaximal oxygen uptake and exercise intensity for discrete ($y = 1.27 + 0.14(x)$; $r = 0.98$) and continuous incremental ($y = 1.92 + 0.12(x)$; $r = 0.98$) submaximal exercise protocols. There were also no differences in the AOD ($\sum$L) when discrete (1.14 ± 2.44) and continuous incremental (0.90 ± 1.12) submaximal exercise protocols were used to determine the supramaximal workload. The oxygen uptake during continuous incremental submaximal exercise tests proved to be reliable over the two testing sessions. The results of Study Two indicated that continuous incremental submaximal exercise protocols are a more time-efficient method of determining the AOD. Study Three compared AOD of pre-pubertal males using running and cycling as the modes of exercise. AOD ($\sum$L) was larger with running exercise (2.25 ± 0.32) when compared with cycling exercise (1.19 ± 0.20) in this sample of pre-pubertal males. The findings of this thesis suggest that AOD identified differences in the anaerobic performance capabilities of children from pre-puberty to post-puberty, and to different exercise

modes. The use of a continuous incremental submaximal exercise protocol has been shown to produce results that were not different from those using discrete submaximal exercise protocols and may make the examination of the anaerobic performance of children using AOD more attractive for pediatric exercise physiologists.

Chapter 1

INTRODUCTION

1.1 Introduction

Accumulated oxygen deficit (AOD) was defined by Green (1994) as an estimation of the total amount of adenosine triphosphate (ATP) resynthesised via anaerobic metabolism by the whole body during short duration high-intensity exercise. AOD is a measure of the difference between the predicted (theoretical) oxygen demand and the actual oxygen uptake for the duration of a supramaximal exercise test (Carlson & Naughton, 1998). AOD has been described as a measure of anaerobic performance and anaerobic capacity (Saltin, 1990; Medbø, et al., 1988; Medbø, 1991; Green and Dawson, 1996b).

AOD has been extensively investigated in adult populations (Bangsbo *et al.*, 1990; Bazdukas *et al.*, 1991; Bangsbo, 1992; Bangsbo *et al.*, 1993; Craig *et al.*, 1993; Gastin & Lawson, 1994a, 1994b; Gastin *et al.*, 1995; Green *et al.*, 1996; Green & Dawson, 1996a; Faina *et al.*, 1997; Buck & Mc Naughton, 1999a; Buck & McNaughton, 1999b; Weber & Schneider, 2000). Studies have shown that AOD is greater in anaerobically trained individuals when compared with untrained adults, (Hermansen & Medbø, 1984; Medbø & Burgers, 1990; Scott *et al.*, 1991; Gastin & Lawson, 1994b) increases with specific anaerobic training, (Medbø, 1991; Tabata *et al.*, 1996) and is higher in males when compared with that of females (Hermansen & Medbø, 1984; Medbø & Burgers, 1990; Scott *et al.*, 1991; Weyand *et al.*, 1993; Gastin & Lawson, 1994b; Weber & Schneider, 2000).

Despite the number of studies reporting the anaerobic performance of adults using AOD, the methodology used to determine AOD remains controversial. The majority of the controversy concerns the assumptions that underpin the methodology used to determine AOD (Green *et al.*, 1996; Bangsbo, 1996a; Graham, 1996a, 1996b). More specifically, the controversy focuses on the use of the relationship between submaximal oxygen uptake and exercise intensity and the extrapolation of that relationship to predict supramaximal energy expenditure. Another controversial issue relates to different methodologies used to determine the relationship between submaximal oxygen uptake and exercise intensity. Different methodologies may be a source of greater errors than those associated with underlying assumptions as inconsistencies make the comparison between studies problematic (Bangsbo, 1996b).

The use of AOD for this series of studies is a measure of performance rather than capacity as AOD is a performance to exhaustion. A limited number of studies have investigated anaerobic performance to exhaustion in children using AOD (Carlson & Naughton, 1993; Buttifant *et al.*, 1996; Naughton *et al.*, 1997). AOD has been described as a potentially viable protocol for use with children due to its "challenging" and non-invasive nature. Naughton et al. (1998) described

AOD as "challenging" because the test was terminated at exhaustion, which is different from other tests of anaerobic performance usually performed by children. However, the controversy that exists when AOD is examined in adult populations does not disappear when used with pediatric populations. Naughton and Carlson (1998) suggested that AOD might be subject to even greater errors in the pediatric population. The authors suggested that it was difficult to obtain a large number of submaximal tests in children because they have a smaller capacity for submaximal exercise when compared with adults. At low submaximal intensities, children may experience an increased oxygen cost in relation to maintaining balance and coordination and, at relatively high intensities, oxygen drift may occur (Naughton and Carlson, 1998). The original proponent of AOD, Jon Medbø (1991), postulated that a large number of steady state oxygen uptake values, at both low and high submaximal intensities, were necessary to achieve an optimal AOD. As a consequence of these observations, Naughton and Carlson (1998) recommended that more research be conducted into the use of AOD with children. Directions for future research encompassed investigations of different testing protocols and the influence of protocols on AOD, the longitudinal and developmental profiles of children's anaerobic performance measured using AOD, the sensitivity of AOD to exercise training interventions in pediatric populations, and changes in the AOD with different exercise modes (Naughton & Carlson, 1998). In light of the recommendations of Naughton and Carlson (1998), the three studies in this thesis have focused on examining the method of determining AOD in the pediatric population.

1.2 Purpose

The studies conducted in this thesis were designed to directly address several of the recommendations made by Naughton and Carlson (1998). In order to do this, a series of three related studies were conducted. The purposes of these studies were:

1. To use accumulated oxygen deficit to measure anaerobic performance in a cross-section of three groups of male children who represented three developmental stages: pre-pubertal, pubertal and post-pubertal.
2. To compare the determination of oxygen uptake at various submaximal exercise intensities using discrete and continuous incremental submaximal running protocols and to examine the repeatability of oxygen uptake during continuous incremental submaximal exercise protocols.
3. To compare accumulated oxygen deficit obtained using treadmill running and cycling as the modes of exercise.

The objective of this series of studies was threefold. Firstly, to quantify the anaerobic performance of children determined by AOD across the maturity continuum from pre-pubertal to post-pubertal The second objective of this series of studies was to examine the method for determining AOD of children in more detail, specifically investigating the impact of different submaximal exercise protocols and finally, to examine AOD in the same group of children using different exercise modes.

1.3 Rationale

The rationale behind this research argues for a need for a greater understanding of the development of anaerobic performance in children by examining a cross-section of maturational development from pre-puberty to post-puberty and quantifying anaerobic performance using AOD. The method for determining AOD will also be examined in greater detail than previously in order to determine whether a less time-consuming methodology can be utilised in the pediatric population. The findings of this research will show the variety of different ways that AOD can be used to quantify the anaerobic performances of children and will meet the need for a laboratory-based test for the examination of the anaerobic performance of children to exhaustion.

1.4 Limitations

In conducting this research, the following limitations are recognised:

1. No attempt was made to alter the lifestyle choices of the children within the framework of this investigation. As a result the following factors were not assessed:

- the physical activity patterns
- dietary choices
- attitudes
- emotional condition, and
- the social environment of the children who participated in the studies.

1.5 Delimitations

1. Concerns over the ethical and moral constraints of research with the pediatric population limited the extent of invasive protocols to blood collection. It was considered unethical and unnecessary to include extremely invasive procedures such as muscle biopsy.

2. A secondary delimitation of the studies was that only the physiological responses to the tests were examined. The sociological and emotional factors associated with the research were not investigated.

3. The research was limited to samples of children from the inner northern and western suburbs of Melbourne. The active nature of the studies delimited them to a sample of volunteers, therefore self-selection of participants may have created a sample bias towards males who willingly participated in moderate to intensive exercise bouts.

1.6 Definition of terms

The following definitions of terms were adopted for use during the study.

Anthropometric measures:

These terms describe the body size of the participants.

Mass (kg) - The total mass of a person.

Height (cm) - The linear size measure of a person.

Cardiorespiratory function measures:

This term describes the measures adopted in this study to reflect the cardiovascular and respiratory function of the children in this study.

Peak oxygen uptake - the highest rate of oxygen use during an incremental test to volitional exhaustion, or the attainment of maximal performance criteria as observed by Zwiren (1989). In this study, peak oxygen uptake tests were predominantly conducted on the treadmill (Studies One and Two). They were conducted on both the treadmill and the cycle ergometer in Study Three. The peak oxygen uptake test imposed incremental loads until volitional fatigue. Peak oxygen uptake is also represented by the symbol $\dot{V}O_2$ (volume of oxygen uptake per minute) and the term $\dot{V}O_2$ max. throughout the thesis. Peak oxygen uptake was expressed in absolute ($L.min^{-1}$) or relative ($mL.kg^{-1}.min^{-1}$) terms.

Steady state - is defined as the point at which equilibrium between the oxygen cost of exercise and the oxygen supply to the exercising muscles becomes apparent (Åstrand and Rodahl, 1986). Steady state is denoted by a plateau in the oxygen cost of exercise ($\dot{V}O_2$) of a single submaximal workload. For the purposes of this thesis, steady state was achieved when the difference in

oxygen uptake ($\dot{V}O_2$) in the final two minutes of the submaximal exercise bout was no more than ± 2 ml $kg^{-1}.min^{-1}$.

RER - the respiratory exchange ratio is the ratio of the volume of expired carbon dioxide to the ratio of measured expired oxygen consumption ($VCO_2/\dot{V}O_2$). As the volume of carbon dioxide increases with effort, RER values increase. Zwiren (1989) reported that values in excess of 1.0 were indicator of maximal performance in children during incremental exercise to fatigue.

Anaerobic performance measures

This term has been adopted to reflect high intensity all-out or constant intensity short-term efforts. **Supramaximal intensity** - short-term exercise conducted at intensity greater than 100% peak oxygen uptake.

Accumulated oxygen deficit - the difference between the predicted (theoretical) oxygen demand and the actual oxygen uptake for the duration of a supramaximal exercise test. Accumulated oxygen deficit is usually presented in the oxygen equivalent of ATP in litres (ΣL) and, or relative to body weight ($\Sigma mL.kg^{-1}$.) (Medbø et al., 1988).

Peak power - the highest mechanical power generated in a short-term high intensity exercise bout over 30 seconds of an all-out cycling test on a cycle ergometer.

Mean power - the average power output over the duration of a 30-second all-out cycle ergometer test. Mean power is usually expressed in absolute (watts) and relative ($watts/kg^{-1}$) terms.

Chapter 2

REVIEW OF LITERATURE

2.1 Introduction

Since the late 1980s, accumulated oxygen deficit (AOD) has been used by a number of researchers to quantify anaerobic performance (Medbø *et al.*, 1988; Medbø, 1991; Bangsbo *et al.*, 1993; Gastin *et al.*, 1995; Green, 1995). There is a considerable body of published literature concerning AOD and its use as a means of quantifying anaerobic performance in adults. In contrast, the published literature relevant to the method of determining AOD of children and quantifying the anaerobic performance in children using AOD is somewhat limited. This review of literature focuses on the published literature examining the method for determining AOD in the adult population and the anaerobic characteristics and performance of children.

Specifically, this review will be divided into two major areas:

- Accumulated oxygen deficit
- Anaerobic characteristics and performance of children

2.2 Accumulated Oxygen Deficit

The following section will focus on AOD as a means of measuring anaerobic performance. The discussion of the AOD will consider its historical context, the practiced methodologies and their inconsistencies, the limitations of the concept, and the assumptions underpinning its measurement. Other physiological and environmental issues that have been described as influencing AOD such as the impact of the active muscle mass, the influence of training and gender differences will also be discussed.

2.2.1 What is accumulated oxygen deficit? - An overview

Green (1994) defined AOD as an estimation of the total amount of ATP resynthesised via anaerobic metabolism by the whole body during short duration high-intensity exercise. AOD is a measure of the difference between the predicted (theoretical) oxygen demand and the actual oxygen uptake for the duration of a supramaximal exercise test (Carlson & Naughton, 1998). AOD was expressed by Medbø et al., (1988) as an amount of oxygen in total litres (ΣL) or relative to body mass as millilitres of oxygen per kilogram (ΣmL.kg^{-1}). AOD has been described as a measure of anaerobic performance (Green & Dawson, 1996b) and anaerobic capacity (Medbø *et al.*, 1988; Saltin, 1990; Medbø, 1991).

2.2.2 *The historical context of accumulated oxygen deficit*

The term "oxygen deficit" was originally discussed by Krogh and Lindhard (1919/20) who described observations of oxygen uptake from the commencement of exercise to the point at which oxygen uptake reached a steady state (Krogh & Lindhard, 1919/20). The term "oxygen deficit" was reintroduced in the late 1960s when it was described as the lag in oxygen uptake in the initial period of exercise during investigations of anaerobic metabolism (Hermansen, 1969). Medbø, (1991) suggested that the results of Hermansen's 1969 study stimulated research interest from the Northern European and Scandinavian scientists who saw the potential of "oxygen deficit" as a means of measuring anaerobic performance. Additional studies conducted during the 1970s (Karlsson & Saltin, 1971; Eriksson *et al.*, 1973; Linnarsson *et al.*, 1974) also used oxygen deficit to quantify anaerobic performance. Hermansen and Medbø (1984) introduced AOD as a means of estimating anaerobic performance from individual responses to submaximal steady state exercise. The authors (Hermansen & Medbø, 1984) used the relationship between submaximal oxygen uptake and exercise intensity determined under submaximal conditions to obtain a least squares linear regression to predict the "oxygen" cost of supramaximal exercise. However, it was not until the research of Medbø et al., (1988) and Medbø and Tabata, (1989) on AOD that more researchers recognised its potential as a means of quantifying anaerobic performance.

2.2.3 *Measurement of accumulated oxygen deficit*

The method proposed by Medbø et al. (1988) for determining AOD required the participant to perform a $\dot{V}O_2$ max test, several submaximal pre-tests and a supramaximal exercise test.

2.2.3.1 Submaximal pre-tests

Medbø et al. (1988) proposed that a series of at least 10 submaximal pre-tests of 10 minutes in duration be conducted. The submaximal pre-tests were conducted at submaximal intensities between 35 - 90%. In order to determine the relationship between submaximal oxygen uptake and exercise intensity, an individual least squares linear regression equation was constructed. The oxygen uptake at each of the exercise intensities could then be regressed against its corresponding exercise intensity. The resultant equation would give the slope and y - intercept of the regression line. A supramaximal workload would then be calculated by using the equation [$y = a + b(x)$] where $y = \dot{V}O_2$ max, and x = workload. Figure 2.1 illustrates this calculation using pediatric data and the treadmill as the mode of exercise.

2.2.3.2 Supramaximal exercise test

Unlike other measures of anaerobic performance, such as the Force-Velocity test or the Wingate Anaerobic test, AOD requires the participant to exercise to exhaustion. When the supramaximal workload has been calculated, an exercise test is conducted. This test is terminated when the participant is exhausted. For example, when the participant is unable to maintain the preset treadmill speed or the revolutions per minute (r.p.m.) decrease below the predetermined rate on the cycle ergometer. AOD is calculated using the oxygen uptake data from expired air collected during the supramaximal test together with supramaximal test time. AOD by definition is the difference between the predicted "theoretical" oxygen demand for the duration of the test subtracted from the actual oxygen uptake for the duration of the test. Figure 2.2 illustrates the calculation of the AOD from actual pediatric data.

Figure 2.1 Sample calculation for supramaximal treadmill speed

"Peter" aged 12.5 years has a mass of 30.04 kg and his peak $\dot{V}O_2$ was 58.87 $ml^{-1}.kg^{-1}.min^{-1}$ on a motorised treadmill.

1. Submaximal testing:

Treadmill Speed	$\dot{V}O_2$ **$mL^{-1}.kg^{-1}.min^{-1}$**
6	33.04
8	45.36
9.1	51.66
10	56.77

The linear regression equation [y = a + b(x)] computed from the above data is:

$\dot{V}O_2$ ($mL^{-1}.kg^{-1}.min^{-1}$) = 0.41 + 5.05(treadmill speed) which has a correlation coefficient of 0.99

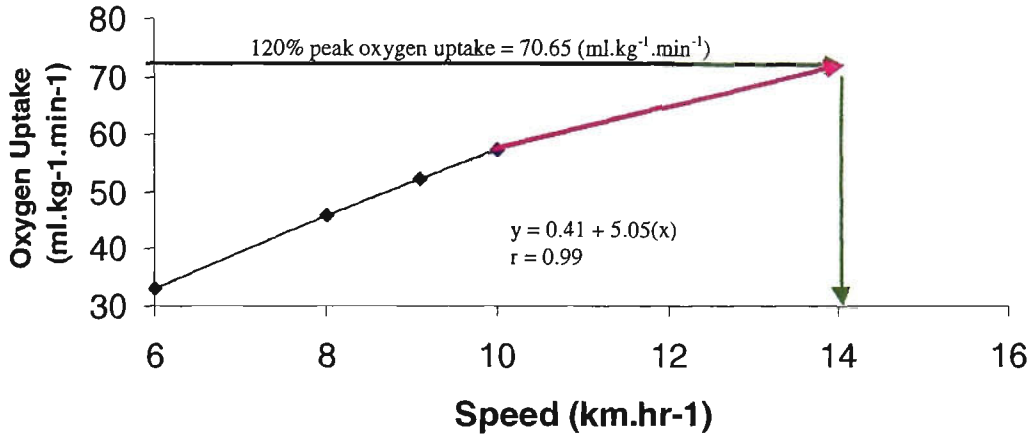
2. Supramaximal prediction

The oxygen uptake at 120% peak $\dot{V}O_2$ is (58.87 x 1.2) = 70.64 $mL^{-1}.kg^{-1}.min^{-1}$. From the linear regression the workload representing 120% peak $\dot{V}O_2$ is calculated.

70.64 = 0.41 + 5.05 (unknown treadmill speed)

Treadmill speed (x) = y – a/b = 13.9 $km.hr^{-1}$.

Figure 2.2 Sample calculation from raw data



Sample calculation of accumulated oxygen deficit

Peter's time to exhaustion at 13.9 $km.hr^{-1}$	= 76.09 s (1268 min)
Total predicted oxygen demand	= 70.65 ($mL.kg^{-1}$ min^{-1}) x 1.268 (min)
for the exercise time	Σ= 89.58 ($ml.kg^{-1}$)

The actual (measured) oxygen consumption for the exercise time from Douglas bag data was 45.96 ($mLkg\text{-}1.min^{-1}$) and for the exercise time= 45.96 ($ml.kg^{-1}$. min^{-1}) x 1.268 (min)

Σ = 58.27 ($ml.kg^{-1}$.)

AOD = Predicted oxygen demand for the exercise time - Actual oxygen consumption for the exercise time

= 89.58 – 58.27
= 31.31 $ml.kg^{-1}$ (relative value)
= 1.04 litres (absolute value)

2.2.4 *Characteristics of accumulated oxygen deficit*

AOD has been proposed as a valid and reliable measurement of anaerobic capacity (Medbø *et al.*, 1988; Medbø & Burgers, 1990; Scott *et al.*, 1991; Gastin *et al.*, 1995). If AOD is justified as a valid and reliable measure of anaerobic capacity, then according to Medbø et al., (1988) it should meet the following criteria:

- it should reach a plateau with increasing exercise duration;
- it should be independent of oxygen uptake;
- it should agree with direct measures of anaerobic metabolism.

2.2.4.1 A plateau with increasing exercise duration

Medbø et al., (1988) investigated AOD in 11 healthy male volunteers. In order to examine the impact of increasing supramaximal test time on AOD, the volunteers were required to participate in treadmill running for a random assignment of supramaximal test times. The test times were 15 sec, 30 sec, 1 min, 2 min and 4 minutes which represented 150%, 130%, 120%, 110% and 105% of peak oxygen uptake, respectively. Treadmill speed was manipulated to bring about exhaustion in the time prescribed for the test; consequently; the treadmill speed was faster during the shorter tests (150 - 130%) when compared with the longer tests (110% - 105%). Exercise was always carried out to exhaustion. Medbø et al., (1988) found that AOD increased ($p<0.001$) with the duration of exercise bouts lasting less than two minutes. When the duration of the test exceeded 2 minutes, a plateau in AOD was observed (Medbø et al., 1988). There were no differences ($p>0.2$) in tests lasting between 2, 4 or 5 minutes. Therefore, the researchers concluded that the AOD had reached a definable maximum for exercise lasting two minutes or longer and as a result could be termed a capacity (Medbø et al., 1988).

A plateau in AOD with increasing exercise intensity has also been observed in the pediatric population by Buttifant et al., (1996) and Naughton et al., (1997). Buttifant et al. (1996) investigated and compared AOD in preadolescent asthmatic and non-asthmatic males using running as the mode of exercise. There was no difference ($p > 0.05$) in the AOD between supramaximal exercise bouts representing 110% and 130% of peak VO_2 in either the non-asthmatic (51.59, 47.04 $mL.kg^{-1}$) or the asthmatic (53.23, 50.60 $mL.kg^{-1}$) males. Buttifant et al., (1996) suggested that the observation of a plateau in AOD with increasing intensity supported the suggestion by Medbø et al., (1988) of an anaerobic capacity. Naughton et al. (1997) examined AOD of male and female adolescent badminton players. Supramaximal exercise bouts that

represented 120 and 130% of peak oxygen uptake were conducted on separate occasions on a motorised treadmill. There were no differences ($p>0.05$) in AOD between the two supramaximal intensities for the male (71.5, 67.6 $mL.kg^{-1}$) or the female (58.6, 58.1 $mL.kg^{-1}$) adolescents. This study also supports the findings of Medbø et al. (1988) of an anaerobic capacity.

2.2.4.2 Independence from maximal oxygen uptake

The independence of AOD from maximal oxygen uptake is supported by the findings of two studies (Linnarsson et al., 1974; Medbø et al., 1988).

The methodology imposed by Linnarsson et al. (1974) on 6 male participants required that they exercise submaximally and maximally at pressures representing hypoxia (0.68 ATA), normoxia (1.00 ATA) and hyperoxia (1.48 ATA). There were no differences in the calculated oxygen deficit (L) measured at maximal intensity and at pressures that represented hypoxia, normoxia and hyperoxia. The oxygen deficit (L) was 5.9 ± 0.3, 5.9 ± 0.4 and 6.3 ± 0.31 for each of the pressures. The main criticism of the study conducted by Linnarsson et al. (1974) was the use of an assumed submaximal efficiency for all participants (Medbø, 1991). Medbø (1991) suggested that individual determination of predicted oxygen deficit costs might have been more precise. Nevertheless, the hypothesis of the study, which focused on the independence of the anaerobic pathways, was supported.

Medbo et al.,(1988) investigated the impact of hypoxia on the $\dot{V}O_2$ max in four participants. The oxygen fraction of the inspired gas was reduced to 13.5 ± 0.2%. The reduction in the inspired oxygen fraction decreased the maximal oxygen uptake by 13 ± 4 $mL.kg^{-1}.min^{-1}$. To examine the effect of hypoxia on AOD, the intensity of the exhausting 4 minute bout was reduced to ~90% peak oxygen uptake. The results indicated an insignificant increase in AOD of 0.7 ± 4.5 $mL.kg^{-1}$ ($p=0.60$). Medbø et al. (1988) concluded that AOD was independent of maximal oxygen uptake.

2.2.4.3 Agreement with direct measures of anaerobic metabolism

Changes in the concentration of the metabolites lactate and phosphocreatine during anaerobic exercise may be used to estimate anaerobic ATP production (Medbø et al., 1988). An observation of the changes in the concentration of metabolites in either muscle or blood during anaerobic exercise has been used to validate AOD (Medbø *et al.*, 1988; Bangsbo *et al.*, 1990). However, blood and muscle metabolites cannot be measured precisely. Methods of measuring metabolites in the blood or in muscle reflect the change in concentration, not the change in the amount of

metabolite. In order to accurately reflect amounts, an assumption of the volumes in which they are distributed must be made. Consequently, the methods of measuring amounts of metabolites yield imprecise results; however, they give an indication of the ability of the participant to generate anaerobic ATP (Medbø et al., 1988).

Estimations of anaerobic pathway activity have been conducted through the measurement of metabolites produced during tests of AOD (Medbø, et al., 1988; Medbø, and Tabata, 1989; Bangsbo, et al., 1990; Bangsbo, et al., 1993; Olesen, 1994; Ramsbottom, et al. 1994; Green and Dawson, 1996b). Bangsbo et al. (1990) investigated AOD during one-legged knee extensor exercise in 8 active male participants. In order to examine the anaerobic energy cost during exercise Bangsbo et al. (1990) required the participants to undergo arterial and venous blood sampling during and post-exercise as well as multiple muscle biopsies from the exercising quadriceps muscles. Blood flow to the rest of the leg was occluded by a pressure cuff placed below the knee. The methodology employed by Bangsbo and his colleagues (1990), although extremely invasive, allowed a direct measurement of the change in metabolites during and post-exercise. The principal finding of this study was that estimates of ATP turnover and anaerobic energy cost were similar to the estimated AOD when direct measures were taken from a single muscle group (Bangsbo et al., 1990).

2.2.4.3.1 Sources of ATP for accumulated Oxygen Deficit

Quantifying anaerobic performance using AOD relies on the precision with which oxygen uptake can be measured. Direct measures of oxygen uptake account for some of the ATP generated during exercise. The energy for ATP in excess of what is accountable for from direct measurement of oxygen uptake is supplied by three means:

1. changes in the O_2 stores of the body, comprised of O_2 bound to hemoglobin and myoglobin, O_2 dissolved in the body fluids and O_2 present in the lungs;
2. the breakdown of PCr and ATP in the exercising muscles; and
3. the breakdown of glycogen to lactic acid, which is partly distributed in the extracellular fluid (ECF).

(Medbø et al., 1988).

By definition, point one is an aerobic source but it cannot be measured directly. If oxygen stored in the body accounts for a large fraction of AOD, then it is a poor estimate of the anaerobic capacity because these stores are aerobic and would be released at the commencement of exercise to facilitate ATP production. Given that AOD is a measure of the oxygen deficit that occurs at the commencement of exercise, large aerobic stores would reduce the need for anaerobic ATP production and therefore reduce the AOD (Medbø et al., 1988). The latter two mechanisms of

anaerobic ATP production have been quantified through invasive research methods such as muscle biopsies and, more recently, non-invasive methods such as magnetic resonance spectroscopy.

The contribution of each source of energy described above to AOD is represented in Table 2.1 below.

Table 2.1 Metabolic components of accumulated oxygen deficit (Adapted from Gastin, 1994)

Component of AOD	Contribution to the AOD mL.kg^{-1} (%)			
	Saltin (1990)*		Medbø et al. (1988)*	Bangsbo et al. (1990)#
	Sed.	AT		
Body stores	5(10)	6(8)	6(9)	(3.3)
Muscle ATP and PCr	15(30)	16 (22)	15.5(24)	(17.4)
Glycolysis and glycogenolysis	30(60)	48(70)	44(67)	(79.3)
Total	**50(100)**	**70(100)**	**65.5(100)**	**(100)**

Sed: Sedentary. AT: anaerobically trained. *Values absolute and (percentage contribution) based on estimates from exercise primarily involving the legs (running or bicycling). #Based on percent contribution observed during exhaustive one-legged knee extensor exercise.

The results presented in the table above are the product of several studies where different methodologies and participants were used. However, collectively the data indicates the contribution of the various components of ATP production to AOD. The data presented in Medbø et al., (1988) and Saltin (1990) are based on predictions of the relative and percentage contributions after measuring the changes in the concentrations of metabolites before and after exercise. The data provided by Bangsbo et al. (1990) comes from actual measurements, taken from muscle biopsy and blood sampling, of the change in the concentration of metabolites after one-legged knee extensor exercise. Bangsbo et al. (1990) examined one muscle group, which explains the smaller contribution from the body stores when compared with the whole body estimations of Medbø et al. (1988) and Saltin (1990). Although Bangsbo and his colleagues (1990) only used one muscle, it was the most accurate means of measuring AOD.

Differences in the percentage contribution from the muscle ATP and PCr stores may be due to the muscles actually involved in the activity. One muscle group is represented in the data of Bangsbo et al. (1990). The source of the data from Medbø et al. (1988) and Saltin (1990) is from running

or cycle ergometer exercise where the ATP and PCr contributions from the muscles would be greater due to the larger muscle mass involved in the exercise.

The figures presented by Medbø et al. (1988) and Saltin (1990) referring to the percentage contribution from glycolysis and glycogenolysis may be subject to some error. The errors may include variations in the dilution of metabolites, differences in the perfusion of the exercising muscle mass and the mode of exercise. The data produced by Bangsbo et al., (1990) which is representative of the knee-extensor muscle, may be closer to what may be expected to be the energy produced from single muscle group. Consequently, the differences in the energy production from glycolysis and glycogenolysis in the studies above may be more difficult to compare.

The data presented by Medbø et al. (1988) and Saltin (1990) which are estimations of the contribution of the various energy systems to AOD, are quite similar. There are differences, however, in the values of the sedentary participants when compared with the trained participants (Saltin, 1990) (Bangsbo *et al.*, 1990). These differences result primarily from the provision of energy from glycolysis and glycogenolysis and suggest that the effect of training on AOD may result from a greater efficiency in the production of energy from glycolysis and glycogenolysis. Gastin (1994) suggested that trained participants did not have much of an advantage over untrained participants in the storage of O_2 in the muscle and in the blood, or in the ability to store ATP and PCr in the muscle. The effect of training and the sensitivity of AOD to training status will be discussed in greater detail later.

2.2.5 Potential sources of error in accumulated oxygen deficit

This section will consider the potential source of errors in AOD.

2.2.5.1 Imprecision of accumulated oxygen deficit according to Medbø

Medbø (1991) systematically addressed the imprecision of AOD. Medbø (1991) considered imprecision to be related to:

- The linearity of the prediction equation.
- The error in predicting supramaximal oxygen demand.
- The duration of the supramaximal exercise bout.
- The estimation of active muscle mass.
- The mechanical efficiency of the participant.

2.2.5.2 The linearity of the prediction equation

As discussed previously (Section 2.2.2.1), the procedure for determining AOD calls for several submaximal pre-tests to be conducted. The submaximal pre-tests establish the relationship between oxygen uptake and exercise intensity. A linear regression equation is then constructed which describes the relationship mathematically. The linear regression equation is then extrapolated to predict a theoretical oxygen demand above $\dot{V}O_2$ max. Medbø (1991) suggested that 90% of the error associated with AOD could be attributed to the imprecision of the estimated oxygen demand from the linear regression equation. The imprecision in the estimation is associated with the scatter around the regression line and the error in the slope of the line (Medbø, 1991).

2.2.5.3 The error in predicting the supramaximal oxygen demand.

The extrapolation of the relationship between submaximal oxygen uptake and exercise intensity that represents supramaximal oxygen demand is not measured. The relationship relies on the linearity of submaximal oxygen uptake and exercise intensity. The linearity of the relationship is subject to errors, which Medbø (1991) discussed. Medbø (1991) was particularly interested in the impact of errors in the extrapolation of the regression line beyond $\dot{V}O_2$ max.

The error in predicting the supramaximal oxygen demand is related to the statistical imprecision in the estimation of the oxygen demand. Random errors of variable size were found in 5-10% of all measurements at submaximal intensities. Medbø et al. (1988) advocated that it was important to try to reduce this error as much as possible. It has been suggested that measures of oxygen uptake at the lowest and highest submaximal intensities will reduce the error in the slope of the regression line. Other sources of error, such as analytical errors in the regression lines, the duration of the supramaximal exercise bout, or the estimation of the active muscle mass may result in the under- or over-prediction of the oxygen demand for the supramaximal exercise bout (Medbø et al., 1988).

2.2.5.3.1 Analytical errors: Regression lines

Medbø et al., (1988) assumed that the errors in the regression line were independent of each other. Variation in measured components of the supramaximal tests contributed to errors in determining AOD. The total variance ($\sum SE^2$) was taken as the sum of the total variance in the measured components and statistical parameters of the supramaximal tests. These components were:

1. oxygen uptake
2. treadmill speed and inclination
3. duration of the test
4. the statistical parameters

5. regression parameters

(Medbø et al., 1988)

The source of imprecision for points 2 and 3 were suggested to be located in the reading of the treadmill speed and inclination and in the duration of the test (Medbø et al. 1988). The effect of the combined imprecision on the calculated AOD in the study conducted by Medbø et al. (1988) was $<1 mL.kg^{-1}$.

Medbo et al. (1988) suggested that the major source of imprecision was in the statistical error of determining the oxygen demand calculated from the linear regression (point 4). The error in estimating the oxygen demand (Sy) is represented by the equation:

$$Sy = [S_{y.x}{}^2 + S_a{}^2 + (X - \chi)^2.S_b{}^2]^{½}$$

Where $S_{y.x}$ is the scatter around the regression line (the estimated standard error of the residual variation not accounted for by the regression of Y on X), and S_a and S_b are the estimated standard errors of a and b (Brown, 1977). The scatter around the regression line ($S_{y.x}$) reflects the variations in oxygen uptake that are independent of the exercise intensity. These errors were of a variable size and were found in 5-10% of all measures of oxygen uptake at submaximal intensities. Medbø et al. (1988) suggested that a visual inspection of the plot of the measurements could be used to exclude the outlying points. The total methodological error in determining AOD in the study conducted by Medbø et al. (1988) was 3 $mL.kg^{-1}$ or 4%.

The extrapolation of the relationship between oxygen uptake and exercise intensity, which represents supramaximal oxygen demand, is not measured. It relies on the linearity of the submaximal relationship. Medbø (1991) discussed the impact of errors in the extrapolation of the regression line beyond $\dot{V}O_2$ max. Medbø (1991) describes the errors as either type a or type b. These errors are represented as S_a and S_b in the Table 2.2 below. Figure 2.3 illustrates the effect of these errors in the extrapolation of the oxygen demand beyond $\dot{V}O_2$ max.

Table 2.2 below outlines the sources of imprecision for AOD as identified in the study conducted by Medbø et al. (1988). The source and magnitude of the errors represented here may differ according to the methodology employed by different researchers and the degree with which methodologies depart from that proposed by Medbø et al. (1988).

Table 2.2 Methodological errors and their influence on the precision of determining AOD (Adapted from Medbø et al., 1988)

Component	Methodological error	Error for two minute bout		Fraction of total variance, %	Relationship of Error to Variation	
		SE, mL.kg	Variance		Exercise Intensity	Duration
O_2 uptake	0.35 $mL.kg^{-1}$ min^{-1}	0.43	0.18	2.0	Independent	Increases
External Work						
Duration	0.005 min	0.45	0.20	2.2	Proportional	Independent
Speed	0.7m/min	0.42	0.18	2.0	Increases by inclination	Proportional
Inclination	0.02°	0.32	0.10	1.1	Proportional to speed	Proportional
Regression Line						
$S_{y.x}$	0.9 $mL.kg^{-1}$ min^{-1}	1.8	3.24	36.0	Independent	Proportional
S_a	0.2 $mL.kg^{-1}$ min^{-1}	0.4	0.16	1.8	Independent	Proportional
S_b	0.0074 $mL.kg^{-1}$ min^{-1}				Independent	
		2.2	4.93	54.9		Proportional
$(X - \chi)$	150 m/min				Increases	
Sum		3.0*	8.99	100.0		

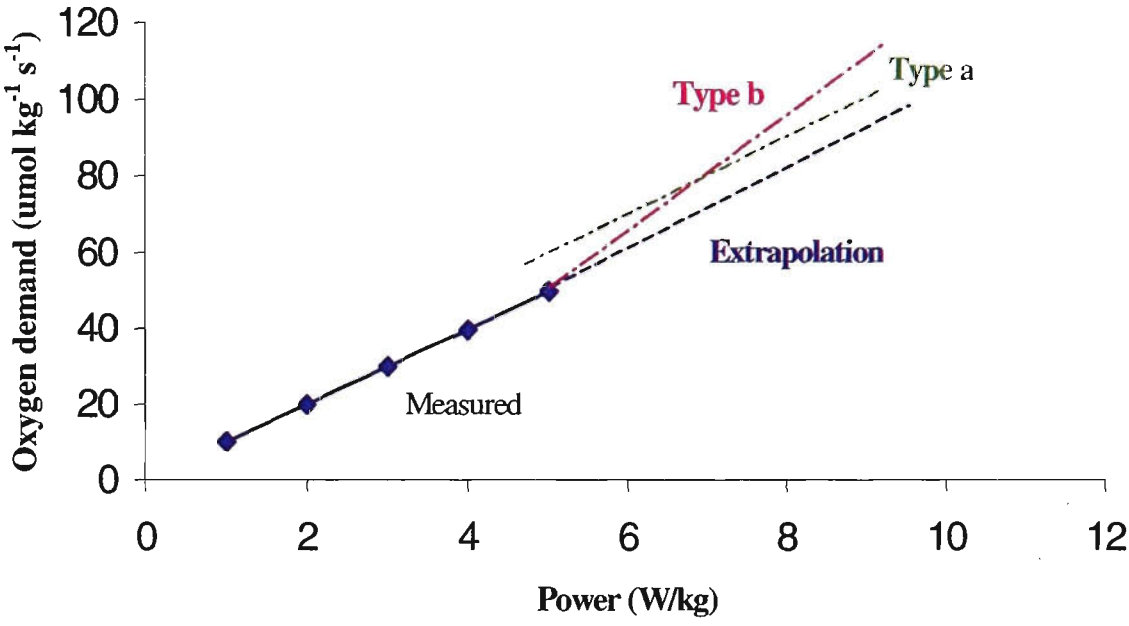
2.2.5.4 Type a error

A type a error is a deviation in the y intercept above the point at which the oxygen demand is extrapolated (the point of discontinuity). The unknown rate of oxygen demand is parallel to the estimated oxygen demand at high intensities. The error is independent of the exercise intensity (Medbø, 1991).

2.2.5.5 Type b error

A type b error is characterised by a deviation in the slope of the regression line that differs from the calculated value. The error increases with the intensity. If AOD determination is to be conducted at a high supramaximal intensity, then the impact of a type b error may be arbitrarily large (Medbø, 1991).

Figure 2.32 Precision of random errors (Adapted from Medbø, 1991)



2.2.5.6 Discussion of type a and type b errors

Medbø (1991) suggested that a true type a error is unlikely. The reality may be that a deviation develops (type b); it reaches a maximum and thus is limited in size. The oxygen demand is a rate, whereas accumulated oxygen demand and AOD are amounts of energy. They are a product of the rate and duration of the supramaximal exercise bout. The effect of either a type a or type b errors are proportional to the duration of the exercise test (Medbø, 1991).

Type a and type b errors are only two of the nonlinear trends that may be possible if the assumed linear relationship between oxygen uptake and exercise intensity is not valid. The relationship between exercise intensity and oxygen demand above the $\dot{V}O_2$ max may be curved. If the relationship is not linear, then it may be explained by internal factors in the muscle or by external mechanical factors (Medbø, 1991).

2.2.5.7 The duration of the supramaximal exercise bout

The duration of the supramaximal exercise bout, whether too long or too short, caused error in the AOD. Medbø (1991) considered that there was an error of approximately 4% for supramaximal exercise lasting less than 2 minutes. Medbø (1996a) identified greater error in tests lasting over 3 minutes. The error is influenced by the statistical imprecision in predicting the supramaximal oxygen demand (Medbø et al. 1988). Medbø (1991) found that the mean absolute difference in AOD between 2 and 4 minutes of exhausting exercise was 0.15 $mmol.kg^{-1}$ or 5%. The expected difference was estimated to be ~ 9%. This was attributed to an error of ~ 4% (2 min) and ~ 7% (4 min) in single determinations of AOD (Medbø, 1991).

Other factors that are thought to influence AOD determination are the day-to-day variations in performance, such as the variation in heart rate and motivation, which may cause participants to perform poorer than they had previously. This may cause variance of up to 5-15% in tests from one day to the next (Medbø, 1991).

2.2.5.8 Estimation of the active muscle mass

Medbø (1991) suggested that an estimation of the active muscle mass during exercise was between 20 and 30% of the body weight for an average adult (male) participant. Active muscle mass was seen as a possible source of error in the AOD as the greater the active muscle mass the higher the AOD.

2.2.5.9 Variations in mechanical efficiency

The relationship between submaximal exercise intensity and oxygen uptake should be determined for each participant individually (Medbø, 1991). Individual differences in the slope of the relationship between oxygen uptake and exercise intensity have been reported as being 6% for treadmill running and 5% for cycle ergometry. The range of differences was 16% and 15% respectively (Medbø, 1991).

Medbø (1991) conducted pilot studies using treadmill running and bicycling as the modes of exercise. The author (Medbø, 1991) observed no important nonlinear effects in oxygen uptake that could be attributed to mechanical differences. Oxygen uptake was observed to be disproportionately large when the participant exercised at low speeds on the treadmill (Medbø, 1991). Greater vertical movement of the participant at low speeds has been suggested as the source of the large oxygen uptake (Medbø, 1991). If these values were included in the linear regression equation, they would have a significant influence on the predicted oxygen demand (Medbø, 1991).

Similar nonlinear effects have been observed on the cycle ergometer at high pedaling frequencies (Medbø, 1991). At frequencies below 1 Hz, no nonlinear effects have been detected (Medbø, 1991). It has been postulated (Medbø, 1991) that the nonlinear effects seen at high frequencies and low powers are the result of imbalances in the body, which necessitate the use of other muscles, such as those in the torso, to maintain balance. The same effects are not apparent at high frequencies and high powers because the reaction forces from the pedals keep the body in balance (Medbø, 1991).

2.2.5.10 Stores of oxygen in the body

At the onset of exercise the total O_2 stores in the muscle, the blood and other body fluids, and in the lungs of an average 77kg (male) participant were estimated to decrease by 450-500ml or 6 $mL.kg^{-1}$ body weight (Medbø, 1991). The main component of this reduction is attributed to the reduction in the O_2 saturation of the mixed venous blood (Medbø, 1991). These oxygen stores contribute to the oxygen deficit at the onset of exercise (Linnarsson et al., 1974), although the quantity of oxygen deficit available from the stores is small compared with the total oxygen deficit (Edwards *et al.*, 1972). The reduction in the O_2 saturation of the mixed venous blood indicates that the O_2 stores contribute little to AOD (Medbø, 1991). Hypoxic experiments have substantiated this by showing no difference in AOD in a limited number of participants (Medbø et al., 1988).

In the transition from rest to exercise, Medbø (1991) identified a delay between the increase in oxygen uptake in the muscles and the point at which it could be measured at the lungs. The stored oxygen is suggested to be equal to ~ 0.25 $mmolO_2$.kg of active muscle mass (Medbø, 1991). Consequently, Medbø (1991) concedes that AOD is a biased measure of the anaerobic energy release. Medbø (1991) reports this error to be ~ 10% in most cases. As the error is related to the proportion of active muscle mass involved in the activity, the mode of exercise and the individual's muscle mass will impact on the magnitude of this error.

2.2.6 The assumptions underlying accumulated oxygen deficit

AOD as proposed by Medbø et al. (1988) requires an acceptance of the assumptions with which the methodology has been validated.

The Assumptions:

- The aerobic energy release can be measured by accumulated oxygen uptake. The anaerobic energy release is the total energy release minus accumulated oxygen uptake.
- The oxygen demand increases linearly with exercise intensity for the type of exercise used.
- The oxygen demand is constant from the onset of exercise if the exercise intensity is kept constant. (Medbø, 1991).

Graham (1996a) suggested that the concept and the theory behind AOD were simple and straightforward. The primary concern, however, are the assumptions that underlie the concept as they are complex and difficult to prove (Graham, 1996a).

Criticism of AOD has been centred on the acceptance of the underlying assumptions (Green, 1995; Green *et al.*, 1996; Bangsbo, 1996a, 1996b; Green & Dawson, 1996b). Medbø (1991) refutes the criticism and suggests that all measures are subjected to random error and are, as a result, imprecise. Indirect measures of the anaerobic energy release during exercise, such as AOD, can never be proven to be accurate. It is possible to detect a large bias accurately; however, small errors may be masked by random variation.

2.2.6.1 Criticism of the assumptions underlying accumulated oxygen deficit

2.2.6.1.1 Oxygen demand increases linearly with intensity

The validity of assumption two, that the oxygen demand increases linearly with exercise intensity for the type of exercise used, has been one of the most strongly criticised assumptions in the literature to date (Bangsbo *et al.*, 1993; Green *et al.*, 1996; Bangsbo, 1996a; Graham, 1996b).

Some authors (Zoladz *et al.*, 1995) suggest that it is difficult to understand why a constant and linear relationship between oxygen uptake and exercise intensity should be expected when the exercise intensity is being increased. Factors such as an increase in the concentration of blood lactate (Casaburi *et al.*, 1987), hydrogen ions (Capelli *et al.*, 1993), catecholamines (Cath, 1971), the recruitment of less efficient type II muscle fibres (Coyle *et al.*, 1992) and increased muscle temperature (Hagberg *et al.*, 1978) have all been proposed as the cause of the slow increase in pulmonary oxygen uptake, also known as oxygen drift, above the resting concentration of blood lactate.

Bangsbo et al., (1993) challenged the assumption of the linearity of oxygen uptake when they investigated AOD in 14 national and international standard runners. The runners produced significantly higher ($p<0.05$) oxygen uptakes at the highest submaximal speed when compared with the relationship between oxygen uptake and exercise intensity at the lower speeds. The authors (Bangsbo et al., 1993) suggested that the relationship between running speed and oxygen uptake during submaximal exercise might not always be linear and may increase disproportionately to exercise intensity when subjects were close to maximum oxygen uptake.

Zolandz et al. (1995) support this concept with results from their study examining the non-linear relationship between oxygen uptake and power output at high submaximal intensities in 12 healthy males participants who exercise on a cycle ergometer. The participants started exercise at 30W with 30W increments imposed until exhaustion. The authors (Zoladz et al., 1995) found a disproportionate increase in oxygen uptake at submaximal exercise intensities above the lactate threshold. Zoladz et al, (1995) also found that actual peak oxygen uptake was approximately 19% lower than that predicted from the relationship between oxygen uptake and exercise intensity calculated during the test. It is not surprising that Zoladz et al. (1995) observed a non-linear relationship between oxygen uptake and exercise intensity because they observed the effects of oxygen drift as a result of the submaximal workloads imposed and the duration of the exercise test.

Criticisms of the findings of Bangsbo et al., (1993) of a non-linear relationship between submaximal oxygen uptake and exercise intensity could be directed at the methodology employed to determine the relationship between submaximal oxygen uptake and exercise intensity. The participants were required to exercise for 6 minutes at each of the submaximal loads. Rest periods of 2 - 10 minutes were allowed between tests. Longer rest periods were taken (up to 10 minutes) as the intensity of the exercise increased. Recent pilot tests suggested that a rest period of at least 30 minutes was necessary to avoid the influence of the excess post-oxygen consumption (EPOC)

of the first test on the oxygen uptake of the second test (Pizza *et al.*, 1996). Ten minutes may not have been sufficient time to reduce the cumulative effect of the EPOC on subsequent tests in the study of Bangsbo et al. (1993). Other investigations evaluating the relationship between oxygen uptake and exercise intensity have used rest periods of 30 minutes (Henson *et al.*, 1989) and 1 -2 hours (Hansen *et al.*, 1988) between tests.

2.2.6.1.2 Extrapolation from submaximal to supramaximal intensities

The criticism of the third assumption of a constant oxygen demand from the onset of exercise if the exercise intensity is kept constant, has been related to the extrapolation of the relationship between submaximal oxygen uptake and exercise intensities to supramaximal exercise intensities (Bangsbo *et al.*, 1990; Maxwell & Nimmo, 1996; Bangsbo, 1996a). The questions concerning the relationship between submaximal oxygen uptake and exercise intensity, changes in mechanical efficiency from submaximal to supramaximal exercise, and the efficiency of the aerobic when compared to the anaerobic production of energy have been raised previously.

2.2.6.1.3 Mechanical efficiency from submaximal to supramaximal exercise intensity

Establishing a relationship between submaximal oxygen uptake and exercise intensity (efficiency relationship) during submaximal exercise, largely accounts for the mechanical inefficiency of an individual participant. The individual efficiency of submaximal exercise can vary by up to 16% for treadmill exercise and 15% for bicycle exercise (Medbø, 1991). If an individual efficiency relationship is not conducted, a group efficiency is adopted; the validity of AOD for an individual must be questioned (Medbø et al., 1988). Concerns about the validity of the assumption that a previously determined submaximal efficiency relationship will carry into a supramaximal efficiency of the same magnitude, have been raised (Bangsbo, 1996b). There are at present, no conclusions with respect to the validity of the extrapolation of the submaximal relationship through to supramaximal exercise. Further studies examining the energy demand during supramaximal exercise have been recommended (Bangsbo, 1996b).

2.2.6.1.4 Contribution of anaerobic and aerobic sources

Accumulated oxygen deficit is noninvasive and allows the supramaximal effort to be easily categorised into aerobic and anaerobic contributions (Medbø & Tabata, 1989). During the supramaximal test, the oxygen uptake is measured and subtracted from the predicted oxygen demand for the duration of the test, thereby enabling the proportional contribution of aerobic and anaerobic energy sources to be quantified. The duration of the test is important when considering the validity of the result. Longer tests will contribute a greater proportion of the energy from

aerobic sources. Medbø (1991) suggested that in tests of 1 minute in duration, the proportional contribution of aerobic and anaerobic energy sources was roughly equal. Medbø (1991) advocated that the test time that produced the smallest error (4%) was a test time between 2 - 3 minutes. The source of the error is in the determination of the predicted oxygen demand. The possibility of a greater contribution from aerobic energy sources, and consequently greater errors, comes from individuals who have a high $\dot{V}O_2$ max, faster O_2 uptake kinetics, and low anaerobic powers (Gastin, 1994). These characteristics are most likely to be found in well-trained endurance athletes. Scott et al. (1991) found a greater aerobic contribution to the supramaximal test in the distance athletes (70%) when compared with the sprinters (61%), middle distance runners (63%) and controls (66%). The authors (Scott et al., 1991) used a common y intercept of 5.0 $mL.kg^{-1}.min^{-1}$ and three 10-minute submaximal tests were performed at 85-100% of $\dot{V}O_2$ max to determine the relationship between submaximal oxygen uptake and running speed of the participants. This protocol may not have been effective in adequately determining the relationship between submaximal oxygen uptake and running speed of the distance group because the distance running group may have been more efficient during submaximal exercise due to their training status (Gastin, 1994). This may have resulted in errors in the determination of the predicted oxygen demand for the supramaximal test.

Another potential source of differences in aerobic contributions of each of the groups in this study (Scott *et al.*, 1991) may be the efficiency with which different muscle fibre types generate and use ATP. Type I muscle fibres have been seen to be more efficient than types IIa or IIb (Coyle *et al.*, 1992; Horowitz *et al.*, 1994). Muscle biopsies of gastrocnemius conducted by Bangsbo et al., (1993) on distance runners (n = 6) revealed fibre type proportions of 78.3 ± 5.6% type I, 20.9 ± 5.8% type IIa and 0.8 ± 0.6% type IIb. Type I fibres have been identified as possessing greater "oxidative" capacity when compared with other fibres (Horowitz et al., 1994). Superior mechanical efficiency has also been found in athletes with a higher percentage of type I fibres (Coyle et al., 1992). The combination of greater oxidative capacity and superior mechanical efficiency will possibly make the determination of the relationship between submaximal oxygen uptake and running speed in endurance athletes subject to greater error if they are not 'pushed' to high enough submaximal work loads. This may have been the case in the study conducted by Scott et al. (1991).

2.2.6.1.5 Criticism of accumulated oxygen deficit

When criticism is directed at the assumptions of AOD, the validity of the assumptions come into question and the measure used to quantify anaerobic capacity is in doubt. The acceptance of the assumptions is critical. The opinions of researchers expressed in the literature either support the assumptions or refute them. This is to a large extent why AOD has not achieved universal acceptance as a measure of anaerobic capacity (Graham, 1996a).

2.2.7 Methodological issues effecting accumulated oxygen deficit

Difficulties in comparing the anaerobic performance of different groups of participants that represent trained, untrained, male and female have been compounded by the fact that most of the researchers have chosen not to follow the procedure proposed by Medbø et al., (1988) (Gastin, 1994). Maxwell and Nimmo (1996) described the methodology proposed by Medbø et al. (1988) as time consuming, involving multiple measures, which are difficult to obtain. The well-controlled procedures of Medbø et al., (1988) "do not readily lend themselves to the more practical and less time-consuming procedures of others" (Gastin & Lawson, 1994a).

2.2.7.1 Different submaximal methodologies

The majority of the time needed to quantify AOD is taken up by the determination of the relationship between submaximal oxygen uptake and exercise intensity (submaximal economy). In response to this time factor, the majority of researchers have simplified the method that they use to determine the submaximal economyy. This simplification has occurred in two ways. The first is by reducing the number of submaximal exercise bouts from 10 proposed by Medbø et al. (1988) to four (Medbø & Burgers, 1990; Pizza *et al.*, 1996), five (Olesen, 1992; Gastin *et al.*, 1995), six (Bangsbo *et al.*, 1993; Weyand *et al.*, 1994; Gastin *et al.*, 1995) and seven (Weyand *et al.*, 1993). The second is by reducing the time of the submaximal exercise bout from 10 minutes proposed by Medbø et al., (1988) to four minutes (Billat *et al.*, 1996; Green & Dawson, 1996b), five minutes (Weyand *et al.*, 1993; Weyand *et al.*, 1994; Gastin *et al.*, 1995), six minutes (Olesen, 1992; Bangsbo *et al.*, 1993; Olesen *et al.*, 1994) and seven minutes (Gastin et al. 1995). More recently, Buck and McNaughton (1999a, 1999b) discussed the impact of reducing the time of the submaximal exercise bouts from the 10 minute bouts, proposed by Medbø et al. (1988), on AOD. The authors (Buck and McNaughton, 1999b) conducted ten 10-minute submaximal exercise bouts to determine the submaximal efficiency. The oxygen uptake during the 10-minute submaximal exercise bouts was then averaged and broken down into two-minute time periods (2 - 4 min, 4 - 6 min, 6 - 8 min and 8 - 10 min). One supramaximal exercise bout (110% $\dot{V}O_2$ max) to determine AOD was conducted using the linear regression equation from the average oxygen uptake from

the 8 - 10 minute time frame. Subsequently, AODs were calculated working back from predicted oxygen demand using 5-second time frames. This study (Buck and McNaughton, 1999b) revealed that AOD calculated from the 2 - 4 minute submaximal economy was significantly different from the estimated AOD from the 8 - 10 minute submaximal economy. All of the other AODs were not significantly different. Buck and McNaughton (1999b) also found no significant differences in regression line parameters or AOD after manipulating the number of data points used in the linear regression equation. The results of this study suggest that submaximal economy relationships for the determination of AOD should be at least 6 minutes in length. Contrary to the suggestions of Buck and McNaughton (1999a, 1999b), Green and Dawson (1996a), suggested that the submaximal test time should be more representative of the supramaximal exercise test time of 3 - 4 minutes. A shorter test time could decrease the role that different stimuli play in influencing ATP turnover (Green & Dawson, 1996a).

2.2.7.1.1 Discrete or continuous submaximal exercise protocols

Medbø et al. (1988) proposed that the submaximal economy relationship be determined during discrete submaximal exercise bouts. Discrete submaximal exercise bouts allow for a rest and recovery break between each submaximal economy determination. The majority of studies have reported that the duration of the rest breaks have been between 3 -10 minutes (Olesen, 1992; Bangsbo *et al.*, 1993; Weyand *et al.*, 1993; Olesen *et al.*, 1994; Faina *et al.*, 1997; Buck & McNaughton, 1999b). One study (Pizza *et al.*, 1996) suggested that rest breaks of 30 minutes between submaximal ecomomy determinations be applied in order to reduce the effect of excess post exercise consumption (EPOC) on the oxygen uptake values of the subsequent submaximal exercise test. Another study (Weber & Schneider, 2000) also allowed 30 minute rest breaks between discrete submaximal exercise test determinations.

In order to decrease the time-consuming nature of discrete submaximal economy determinations, a number of authors have used continuous incremental submaximal exercise determinations (Graham & McLellan, 1989; Bangsbo *et al.*, 1990; Craig *et al.*, 1993; Green & Dawson, 1996a; Jacobs *et al.*, 1997). The continuous incremental exercise bouts have been between 4 and 8 minutes in duration and between 3 - 9 determinations have been conducted in one continuous incremental exercise bout. Medbø's earlier studies on AOD proposed discrete determinations of the submaximal economy relationship (Medbø et al., 1988). However, he later supported the use of continuous incremental protocols and suggested that each increment should be between 5 - 8% of the participant's $\dot{V}O_2$ max every 4 minutes (Medbø, 1996b).

2.2.7.1.2 Common y-intercept in the regression equation to predict supramaximal intensity.

Several researchers have used a common y-intercept (5 $mL.kg^{-1}. min^{-1}$) in the regression equation used to predict the supramaximal exercise intensity (Scott *et al.*, 1991; Ramsbottom *et al.*, 1994; Maxwell & Nimmo, 1996; Faina *et al.*, 1997; Wadley & Le Rossignol, 1998). However, Medbø et al. (1988) supported the use of a common y-intercept, and there have been few reports of large deviations from the 5 $mL.kg^{-1}. min^{-1}$ resting oxygen uptake. The validity of using a common Y-intercept is still subject to some concern as the sample size in the study was limited to 11 (Medbø et al., 1988) and individual variability may be greater in a larger sample size (Gastin, 1994).

2.2.7.1.3 Comparing the submaximal efficiency determinations from discrete and continuous submaximal protocols.

To date, only one study has investigated the differences in the determination of the submaximal economy relationship from both discrete and continuous incremental submaximal exercise protocols (Green & Dawson, 1996a). Green and Dawson, (1996a) found no significant differences in the submaximal economy relationships when the discrete and continuous incremental submaximal exercise protocols were compared. The authors (Green and Dawson 1996a) did, however, observe a slight increase in oxygen uptake towards the end of the continuous incremental exercise test, but could not suggest what the mechanisms of the increase in oxygen uptake might be.

2.2.7.1.4 Section summary

A variety of methodologies other than that proposed by Medbø et al., (1988) have been employed by researchers to determine the submaximal ecomomy relationship. These methodologies have included decreasing the number of submaximal exercise bouts, decreasing the duration of submaximal exercise bouts and using continuous incremental exercise bouts as opposed to discrete submaximal exercise bouts. The reasoning behind the differing methodologies has been largely to decrease the time-consuming nature of the methodology proposed by Medbø et al., (1988). Buck and McNaughton (1999b) summed up the position when they suggested that further research was required into AOD that included a standardisation of the submaximal exercise protocol.

2.2.7.2 Supramaximal exercise bouts

2.2.7.2.1 Constant intensity supramaximal exercise bouts

The majority of researchers conduct supramaximal exercise bouts at a constant intensity that is predicted from the linear regression equation represening the submaximal efficiency relationship (Medbø *et al.*, 1988; Graham & McLellan, 1989; Medbø & Tabata, 1989; Medbø & Burgers, 1990; Olesen, 1992; Bangsbo *et al.*, 1993; Olesen *et al.*, 1994; Jacobs *et al.*, 1997; Buck & McNaughton, 1999b; Weber & Schneider, 2000). These supramaximal tests have been conducted at intensities that represent 110 - 140% of $\dot{V}O_2$ max to exhaustion. The criterion for exhaustion has been a failure of the participants to maintain a pedal cadence above 60 r.p.m. on the cycle ergometer or to maintain the treadmill speed.

2.2.7.2.2 All-out supramaximal exercise bouts

Some researchers have used all-out supramaximal tests for a pre-determined time period to measure AOD (Withers *et al.*, 1991; Withers *et al.*, 1993; Gastin & Lawson, 1994a, 1994b; Gastin *et al.*, 1995; Hargreaves *et al.*, 1997). Pre-determined exercise times during all-out tests have varied between 45 - 90 seconds. Gastin and Lawson (1994b) suggested that all-out tests might be an alternative to constant intensity tests, as they appear to be reliable. Gastin et al., (1995) compared AOD after constant intensity and all-out tests. The authors (Gastin et al., 1995) found that the all-out test was a valid estimate of AOD.

Medbø et al. (1988) suggested that supramaximal tests used to measure AOD be continued until the participant was exhausted. Exercise to exhaustion was also one of the criteria for determining that AOD was a measure of anaerobic capacity. All-out tests are conducted for a researcher-defined period of time and therefore may not completely exhaust the anaerobic capacity of the participant.

2.2.7.2.3 Measuring oxygen uptake during supramaximal exercise bouts

Medbø et al. (1988) suggested that oxygen uptake during supramaximal exercise be measured using Douglas bags. The advantage of using Douglas bags is that all of the expired oxygen can be accounted for and be used to measure the oxygen uptake during the supramaximal exercise bout. Some researchers (Graham & McLellan, 1989; Bangsbo *et al.*, 1990; Scott *et al.*, 1991; Green & Dawson, 1996a; Faina *et al.*, 1997; Buck & Mc Naughton, 1999a; Buck & McNaughton, 1999b) have used on-line metabolic systems to measure oxygen uptake during the supramaximal exercise tests. The oxygen uptake measured during these tests may be underestimated if the participant has not been able to complete the sampling time frame determined by the on-line metabolic system. Green et al. (1996) measured oxygen uptake during the supramaximal tests using an on-line system that calculated oxygen uptake at 15-second intervals. The authors (Green et al., 1996)

attempted to overcome the problem of the participants completing their test prior to a 15-second time interval ending by taking the oxygen uptake from the previous two periods and multiplying it by the time in the final period as a fraction of 15 seconds. This method may be time consuming and not as accurate as collecting expired gases in Douglas bags. Medbø (1991) suggested that the errors associated with measuring oxygen uptake would be reduced when manual high precision equipment such as Douglas bags was used. Medbø (1996a) also observed that on-line systems only produced average oxygen uptake values and that this was a limitation of their use.

2.2.7.2.4 Section summary

Supramaximal tests have been conducted using constant exercise intensities and all-out supramaximal exercise for a researcher-determined time frame. Oxygen uptake during the supramaximal tests has also been measured using Douglas bags and on-line metabolic systems. Medbø et al., (1988) proposed that in order to reduce the errors in determining AOD from supramaximal tests the supramaximal tests should be conducted at a constant intensity to exhaustion and oxygen uptake should be measured using Douglas Bags as opposed to on-line metabolic systems.

2.2.8 The reliability of accumulated oxygen deficit

AOD has been demonstrated to be a reliable measure (Lawson & Golding, 1981; Graham & McLellan, 1989; Carlson & Naughton, 1993; Ramsbottom *et al.*, 1994; Jacobs *et al.*, 1997). Carlson and Naughton, (1993) reported test re-test reliability of 0.94 in nine pre-pubertal males and 0.57 in nine pre-pubertal females. The authors (Carlson and Naughton, 1993) suggested that the difference in the test re-test reliability of the pre-pubertal girls reflected poorly on their performances when compared with those of the pre-pubertal boys. In adults, Graham and McLellan (1989) found a coefficient of variation in AOD of 8 - 13% in four trained cyclists. Jacobs et al. (1996) reported correlation coefficients of 0.94 between two supramaximal exercise tests prior to a creatine monohydrate intervention study in a sample of fourteen participants. In all-out supramaximal exercise bouts, Lawson and Golding (1981) reported correlation coefficients of 0.98 between tests. Ramsbottom et al. (1994) found a correlation coefficient ($r = 0.94$) between two different tests at the same supramaximal intensity in a group of 12 trained runners.

Gastin (1994) suggests that the problems with obtaining reliable measures of AOD may lie with the accurate determination of the time to exhaustion in constant intensity tests and motivational problems that may be apparent during longer exercise durations. The reliability measures of the

supramaximal test are related to the repeatability of a supramaximal test. They do not differentiate between technological or biological variability nor do they take into account the reliability of the submaximal efficiency relationships that are used to predict the supramaximal workload (Gastin, 1994).

2.2.9 Validity of accumulated oxygen deficit

The validity of AOD can be considered from a number of different perspectives. Firstly, AOD is by definition the difference between the predicted oxygen demand (oxygen cost) and the aerobic contribution (oxygen uptake) during a supramaximal exercise test. The energy for a supramaximal exercise test comes from anaerobic and aerobic sources. The energy released during a supramaximal exercise test is the sum of two components: an aerobic component that is proportional to the duration of the supramaximal test, and a constant anaerobic component (Gastin, 1994). This point is supported by numerous studies that have found that AOD reaches a plateau with increasing supramaximal test durations(Medbø *et al.*, 1988; Buttifant *et al.*, 1996). AOD has also been shown not to change in hypoxic conditions (Linnarsson *et al.*, 1974; Medbø *et al.*, 1988).

Secondly, AOD has been reported to be strongly correlated with direct measures of anaerobic metabolism such as muscle biopsies (Bangsbo *et al.*, 1990; Medbø & Tabata, 1993). Contrary to these findings, Green et al. (1996) found no relationship between AOD and anaerobic ATP production or enzyme activities in 10 trained cyclists. To date, this has been the only study that has refuted the suggestion that there is a strong relationship between AOD and direct measures of anaerobic metabolism.

Unfortunately, anaerobic metabolism measured from muscle biopsy has its limitations and validating AOD against muscle biopsies also relies on the validity of a number of assumptions relating to muscle biopsies. Despite these limitations, and because there is no direct measure of anaerobic capacity, unlike aerobic capacity, the comparison with muscle biopsies remains the best method of validating AOD (Gastin, 1994).

Thirdly, AOD has been shown to respond to anaerobic training (Medbø, 1991). Medbø and Burgers (1990) reported a 10% increase in AOD after six weeks of anaerobic training in a group of five males and seven females. Heugas et al. (1997) investigated the response of AOD to intense aerobic training in a group of elite 400m male sprinters (n = 11). The authors (Heugas et al., 1997) observed a decrease in AOD from 64.9 ± 10.8 $mL.kg^{-1}.min^{-1}$. to 50.2 ± 9.2 $mL.kg^{-1}.min^{-1}$ ($p<0.05$) after three months of aerobic training. A 8.86% increase in $\dot{V}O_2$ max was also

observed during the training period (Heugas *et al.*, 1997). Heugas et al. (1997) concluded that the AOD was sensitive to intense aerobic training in elite male 400m sprinters.

Jacobs et al. (1997) investigated the effect of the ingestion of creatine monohydrate on anaerobic performance measured using AOD. A total of 26 participants were randomly assigned to double blind trials of a placebo (n = 12) or creatine monohydrate (n = 14) ingestion for 5 days. Supramaximal tests to determine AOD were performed twice prior to the treatment and twice post-treatment (Jacobs *et al.*, 1997). The authors (Jacobs et al., 1997) reported an increase in AOD from 4.04 ± 0.31 to 4.41 ± 0.34 (L)($p<0.001$) in the group that ingested creatine monohydrate for 5 days.

Fourthly, the validity of AOD as a measure of anaerobic performance suggests that there should be a relationship between AOD and athletic performances that are known to demand a high anaerobic component. Ramsbottom et al. (1994) examined the relationship between AOD and track running performances over 100, 400m, and 800m. The authors, (Ramsbottom et al., 1994) found strong correlations between 100m ($r = -0.88$, $p<0.01$) and 400m ($r = -0.82$, $p<0.01$) track times and measures of AOD. A strong correlation was not observed between 800m running time and AOD ($r = -0.61$, $p<0.05$). Ramsbottom et al. (1994) concluded that AOD was a valid measure of anaerobic performance.

Scott et al. (1991) also reported a strong correlation between 300m run time and AOD ($r = -0.76$, $p<0.01$). AOD was also strongly correlated with other measures of anaerobic performance such as peak power during the Wingate Anaerobic Test ($r = 0.70$, $p <0.01$). The authors (Scott et al., 1991) concluded that correlations between AOD and other predominantly anaerobic work tests supported the validity of AOD as a means of measuring anaerobic performance.

Gastin (1994) questions the validity of AOD in light of the fact that the AOD is an estimation of the energy cost; it is not an actual measurement. AOD relies on an assumption that the mechanical efficiency of submaximal exercise does not change when extrapolated to supramaximal work. Despite the questionable validity of the submaximal to supramaximal efficiency assumption, AOD still remains a promising means of quantifying the anaerobic capacity in a non-invasive protocol (Gastin, 1994).

2.2.10 Physiological influences on accumulated oxygen deficit

The magnitude of AOD appears to depend on the anaerobic training status, the genetic predisposition of the individual to anaerobic exercise, and the size of the muscle mass involved in

the exercise. Tables 2.3 - 2.5 present a summary of AOD to date using a variety of exercise protocols, exercise modes and participants.

Table 2.3 Summary of AOD using cycling as the mode of exercise

Reference	Training status	n	Submaximal method	AOD L ($mL.kg^{-1}$)
(Eriksson *et al.*, 1971)	UT	3	AME	3.8(~55)
(Linnarsson *et al.*, 1974)	UT	6	AME	5.9(~79)
(Davis *et al.*, 1981)	T	5	GLE	4.44(~56)
(Lawson & Golding, 1981)	UT	20	AME	3.39(46)
(Pate *et al.*, 1983)	UT	10	Unknown	4.05
(Graham & McLellan, 1989)	T	4	Cont., S, M, ILR	4.38(~60)
(Medbø & Burgers, 1990)	UT	17	Disc., L, H,ILR	3.12(~42) 3.78(~50) 4.07(~54)
(Bangsbo *et al.*, 1993)	T	3	Disc., I, M-H, ILR	~4.49(56.5)
(Withers *et al.*, 1993)	T	6	Disc., L, L, ILR	3.52(~48)# 3.75(51.0) 3.80(51.9) 3.75(51.1)
(Gastin & Lawson, 1994a)	UT ET ST	8 8 8	Disc., I, M, ILR	3.52(50)# 3.82(52) 4.82(60)
(Gastin & Lawson, 1994b)	UT	8	Cont., I, M, ILR	3.48(47.6)# 3.58(49.0) 3.63(49.6)
(Green & Dawson, 1996a)	T	10	Cont., L, M - H, ILR	4.11(55.2)
(Pizza *et al.*, 1996)	UT RT ET	12 11 10	Disc., L., L, ILR Disc., L., L, ILR	3.07(38.8) 4.75(52.5) 3.7(53.7)
(Tabata *et al.*, 1996)	T	7	Disc., L, H, ILR	~4.7(69.1)
(Faina *et al.*, 1997)	T	8	Disc., I, M, ILR	~3.4(43.6)
(Hargreaves *et al.*, 1997)	T	9	Disc, S, L, ILR	~4.3(~54.5)
(Buck & Mc Naughton, 1999a)	T	8	Disc, L, H, ILR	~3.7(53.4)
(Weber & Schneider, 2000)	UT	10M 10F	Disc, L, H, ILR	3.6(46.3) 2.4(38.2)

Male participants unless indicated. UT: untrained. T: trained (unspecified). ET: endurance trained. RT: resistance trained. Disc.: discrete. Cont.: continuous incremental. S: short (3 -4 min). I: intermediate (4 -6 min). L: long (8 - 10 min). L: low (~4 points used in linear regression). M: medium (~6 points used in linear regression). H: high (~10 points used in linear regression). AME: assumed mechanical efficiency. ILR: individual linear regression. GLE: group linear extrapolation. ~ Approximate calculated values based on mean body weights. # All-out exercise as opposed to constant intensity exhaustive exercise.

Table 2.4 Summary of AOD using running as the mode of exercise

Reference:	Training status	n	Submaximal method	AOD L ($mL.kg^{-1}$)
(Hermansen & Medbø, 1984)	ET	6	Disc., L, L, ILR	~2.9(42)
	ST	6		~3.41(56)
(Medbø & Burgers, 1990)	UT	4	Disc., L, H, ILR	4.72(~64)
	T (ET & ST)	7		6.04(~77)
(Medbø & Burgers, 1990)	UT	6	Disc., L, L, ILR	~4.87(64)
	ET	6		~4.42(64)
	ST	8		~6.30(83)
(Scott *et al.*, 1991)	UT	4	Disc., L, L, ILR	~4.63(56.1)
	ST - MD	8		~5.49(76)
	ET	4		~3.78(56.9)
(Bangsbo *et al.*, 1993)	T(Soccer)	15	Disc., I, M-H, ILR	~3.94(49.5)
	T(Runners)	14		~3.62(51.9)
	T (Rowing)	5		3.65(47.3)
(Ramsbottom *et al.*, 1994)	T	11M	Disc., L, L, ILR	4.62(65.2)
		1F		
(Maxwell & Nimmo, 1996)	T	18	Disc., L, L, ILR	~5.58(74.6)

Male participants unless indicated. UT: untrained. T: trained (unspecified). ET: endurance trained. Disc.: discrete. Cont.: continuous incremental. S: short (3 -4 min). I: intermediate (4 -6 min). L: long (8 - 10 min). L: low (~4 points used in linear regression (L.R)). M: medium (~6 points used in LR). H: high (~10 points LR). ILR: individual linear regression. ~ Approximate calculated values based on mean body weights.

Table 2.5 Summary of AOD using a variety of submaximal exercise protocols and exercise modes

Reference:	Training status/ Exercise mode	n	Submaximal method	AOD L ($mL.kg^{-1}$)
(Bazdukas *et al.*, 1991)	Trained Freestyle	13	Disc., L, ILR	54(52KJ)
	Trained Breaststroke/Swimming			64(58KJ)
(Terrados *et al.*, 1991)	Trained (kayak)/Kayaking	12	unknown	45.91
(Bazdukas *et al.*, 1991)	ET (swim)/Swimming	6	unknown	~2.6(52KJ)
	ST (swim)/Swimming	6		~2.8(56KJ)
(Bangsbo *et al.*, 1993)	T(row)/Rowing	5	Disc., I, M-H, ILR	~4.94(64.1)
(Faina *et al.*, 1997)	T(swim)/Swimming	8	Disc., I, M, ILR	~4.23(56.4)
	T(kayak)/Kayaking	7		~2.97(40.2)

Male participants unless indicated. UT: untrained. T: trained (unspecified). ET: endurance trained. Disc.: discrete. Cont.: continuous incremental. S: short (3 -4 min). I: intermediate (4 -6 min). L: long (8 - 10 min). L: low (~4 points used in linear regression (L.R)). M: medium (~6 points used in LR). H: high (~10 points LR). ILR: individual linear regression. ~ Approximate calculated values based on mean body weights.

2.2.10.1.1 The mode of exercise used to determine accumulated oxygen deficit

AOD has been determined using the cycle ergometer, the motorised treadmill, swimming, kayaking and rowing. Tables 2.3, 2.4 and 2.5 present the range of AOD values published to date. Although Graham (1996b) cautioned against the comparison of AOD values reported in studies with differing methodologies, a comparison of studies provides an overall picture of AOD using different modes of exercise, participants of differing training status, and genders. Gastin (1994) also cautioned against the use of studies that used an assumed mechanical efficiency to predict the supramaximal workload. These studies have been presented in the tables above but have not been used in the discussion of the differences in AOD.

General observations from the tables above indicate that tests conducted on the treadmill (Medbø *et al.*, 1988; Medbø, 1991; Scott *et al.*, 1991) produce a larger AOD than those that have used a cycle ergometer (Medbø & Tabata, 1989; Pizza *et al.*, 1996; Weber & Schneider, 2000) in untrained participants. On the cycle ergometer, untrained participants tend to have smaller reported AODs (Medbø & Tabata, 1989; Pizza *et al.*, 1996; Weber & Schneider, 2000) when compared with endurance trained (Pizza *et al.*, 1996) and sprint trained athletes (Green & Dawson, 1996a). On the treadmill, the same trend in performances is observed with untrained participants (Medbø *et al.*, 1988; Scott *et al.*, 1991) producing a smaller AOD than those reported for sprint trained athletes (Medbø *et al.*, 1988; Scott *et al.*, 1991; Ramsbottom *et al.*, 1994; Maxwell & Nimmo, 1996). Endurance trained athletes (Hermansen & Medbø, 1984; Medbø & Burgers, 1990; Scott *et al.*, 1991) produce smaller AODs that those of untrained (Medbø and Burgers, 1990; Scott et al., 1991) and sprint trained participants (Medbø and Burgers, 1990; Scott et al. 1991). These findings suggest that AOD is sensitive to differences in training status (Medbø and Burgers 1990; Scott, et al., 1991; Tabata, et al., 1996; Heugas, et al., 1997).

2.2.10.2 Muscle mass involved in exercise: Impact on accumulated oxygen deficit

Medbø (1991) quantified AOD for running and cycling exercise but not for swimming and kayaking exercise. Consequently, the extrapolation of the relationship between submaximal efficiency and supramaximal exercise, in studies using swimming and kayaking as the mode of exercise, may be subject to greater errors because the submaximal efficiency relationship has not been examined to the same extent. Table 2.5 presents AOD values for exercise other than cycling or running. The results from these studies suggest that the greater the muscle mass engaged in exercise, the greater we can expect AOD to be. Kayaking (Terrados *et al.*, 1991; Faina *et al.*,

1997) produces smaller AODs than does swimming (Bazdukas *et al.*, 1991; Faina *et al.*, 1997) and rowing (Bangsbo et al., 1990).

Some researchers have discussed the impact of the size of the active muscle mass on AOD (Bangsbo et al., 1993; Weyand et al. 1993; Weber and Schneider, 2000). Bangsbo et al. (1993) determined the submaximal efficiencies and conducted supramaximal tests with trained rowers, running on a treadmill and rowing on a rowing ergometer. The authors (Bangsbo et al., 1993) found that the rowers produced a higher AOD on the rowing ergometer (~4.9L) than on the treadmill (~3.6L). Bangsbo et al., (1993) suggested that the larger muscle mass involved in rowing exercise increased AOD.

Weyand et al. (1993) investigated whether there was a difference in AOD between one-legged and two-legged cycling. The authors (Weyand et al., 1993) reported that AOD was higher after two-legged cycle exercise (4.40L) than one-legged cycle exercise (2.27L). This study supported the hypothesis that AOD was dependent on the size of the active muscle mass (Weyand et al., 1993).

Another study investigating the impact of treadmill gradient on AOD observed a larger AOD when the gradient of the treadmill was raised from 1% (39.5 $mL.kg^{-1}$) to 15% (71.7$mL.kg^{-1}$). No further increase was observed in AOD when the treadmill gradient was increased to 20% (69.4$mL.kg^{-1}$) (Olsen, 1992). Olesen (1992) suggested the increase in the size of the muscle mass engaged in the activity was a mechanism for the doubling in AOD from 1% treadmill gradient to 15-20% treadmill gradient.

2.2.10.3 The impact of training on accumulated oxygen deficit

A limited number of training studies have been conducted to evaluate the impact of training on AOD (Medbø and Burgers 1990; Troup, et al., 1991; Tabata, et al., 1996; Heugas, et al., 1997). Medbø and Burgers (1990) have produced the most comprehensive training study to date. They (Medbø and Burgers, 1990) investigated the impact of two different types of anaerobic training on a group of 5 untrained men and 7 untrained women. Group A consisted of 3 males and 3 females who participated in 40 ± 5 days of training designed to stress their anaerobic capacity. Group A trained at ~115% of their $\dot{V}O_2$ max in three sessions which consisted of three 2-minute efforts. Group B comprised of 2 males and 4 females. Group B undertook training that was designed to stress a high rate of anaerobic energy release (anaerobic power) for the same total time frame (including recovery) as Group A. Each training session consisted of eight repeated 20

second efforts at ~165% of their $\dot{V}O_2$ max. Post-training results revealed an increase in AOD of 10% for both groups. There were no differences in the results of Group A and Group B. Medbø and Burgers (1990) suggested that the different anaerobic training programs did not have an effect on AOD.

Tabata et al. (1996) examined the effect on AOD of 6 weeks of training 5 days per week. Four out of the five days' training was described as exhaustive intermittent training on the cycle ergometer. The participants were encouraged to complete seven to eight sets of the prescribed intermittent training in a training session. When the participants were able to complete nine sets, the resistance on the cycle ergometer was increased by 11 watts. In the fifth training session, the participants completed 30 minutes of cycling at 70%of their $\dot{V}O_2$ max and then completed four sets of intermittent exercise at 170% $\dot{V}O_2$ max. AOD was measured prior to training, at 4 weeks of training, and post 6 weeks training. Tabata et al. (1996) observed a 23% increase in AOD up to 4 weeks of training and a further 5% increase from 4 weeks to 28% increase post training.

In contrast to studies examining the impact of anaerobic training on AOD, Heugas et al. (1997) reported that AOD decreased from 64.9 ± 10.8 $mL.kg^{-1}.min^{-1}$. to 50.2 ± 9.2 $mL.kg^{-1}.min^{-1}$ ($p<0.05$) after three months of aerobic training. The authors (Heugas et al., 1997) also observed an 8.86% increase in $\dot{V}O_2$ max during the training period.

2.2.10.4 Gender differences in accumulated oxygen deficit

Gender differences in the magnitude of AOD indicate that AOD is greater in men when compared with that in women (Medbø and Burgers 1990; Weyand, et al., 1993; Weber and Schneider, 2000).

Medbø and Burgers (1990) observed a smaller AOD in the females involved in their training study when compared with the males. They also observed a smaller increase in AOD in the female group when compared with the male group after 6 weeks of training. Medbø and Burgers (1990) suggested that the anaerobic capacity might be more trainable for men than for women.

Weyand et al. (1993) found that AOD values for males during one-legged and two-legged cycling were greater than that for females. The observed differences in AOD were greater in this study (Weyand et al., 1993) when compared with those of Medbø and Burgers (1990), even although the body mass of the participants in the two studies was similar. The authors (Weyand et al.,

1993) suggested that this might be due to genetic factors, such as muscle fibre type composition or behavioral factors such as the state of training.

Weber and Schneider (2000) investigated AOD in untrained males and females and estimated active muscle mass (AMM) for cycling using dual-energy x-ray absorptiometry (DEXA). They reported a higher AOD for the male participants (126.3 $mL.kg^{-1}.AMM^{-1}$) when compared with the female participants (108.3 $mL.kg^{-1}AMM^{-1}$). Weber and Schneider (2000) suggested that, because there were still gender differences in AOD after they were corrected for estimated active muscle mass, there may be physiological, biochemical and/or structural differences in the muscle tissue of males when compared with that of females which impacted on AOD.

2.2.10.5 Section summary

Studies investigating AOD to date have found that it is related to the size of the active muscle mass engaged during exercise. These findings are supported by findings of a larger AOD from running exercise when compared with cycling exercise. In the same group of participants, AOD was larger when rowing exercise was compared with running exercise (Bangsbo et al., 1990). Training status, training studies and gender have all been shown to influence AOD.

2.3 The anaerobic characteristics and performance of children

The following section will focus on the anaerobic characteristics and performance of children. The anaerobic characteristics of children and their subsequent anaerobic performances are difficult to separate because the characteristics ultimately govern the performance. The anaerobic characteristics of children are different from those of adults and show a developmental trend from pre-puberty to the attainment of adult characteristics post-puberty.

2.3.1 The anaerobic characteristics of children

Maximal oxygen uptake that is readily measured in a laboratory setting has been considered to be a definitive measure of aerobic performance. Maximal oxygen uptake is well described in adult and pediatric populations; this is largely due to the fact that there is a standardised test that makes good use of the scientist's ability to measured expired oxygen. In contrast, anaerobic performance does not have a standardised test and is less well described, especially in the pediatric population. This juxtaposition may well exist because the health benefits of aerobic performance and its relationship to cardiovascular disease may make it more attractive as a research option in comparison with the performance orientation of anaerobic performance (Van Praagh, 2000). Many authors suggest that the pediatric population, who spend the majority of their play and recreation activities participating in activities that require short bursts of activity followed by rest, rely more heavily on a supply of energy from anaerobic means than aerobic means (Roemmich & Rogol, 1995; Rowland, 1996; Van Praagh, 2000; Van Praagh, 2002). Sargeant (1989) suggested that short-term high intensity exercise tests are good tests of functional capacity in children, because they are similar to their habitual activity patterns in their intensity and duration.

Even though children rely on energy supplied by anaerobic means for their play and recreation activities, there is a comparative dearth of published research, when compared to that of aerobic performance, in relation to their characteristics and performance (Roemmich & Rogol, 1995). The few studies that have been conducted have used different tests, of different durations, methodologies and limb involvement (Inbar & Bar-Or, 1986; Blimkie *et al.*, 1988; Vandewalle *et al.*, 1989; Van Praagh *et al.*, 1990; Delgado *et al.*, 1993; Suei *et al.*, 1998,). Given the limited research in this area, studies have shown that there is a developmental increase in anaerobic performance in children. The anaerobic performances of male children are greater when compared with those of females, and males reach their peak later than females (Van Praagh, 2000). Observations of an increase in anaerobic performance are also found when anaerobic performance is allometrically adjusted for body size. This indicates that anaerobic performance in children increases at a greater rate than that which can be ascribed to growth alone (Rowland,

1996). Short term high intensity tests are good tests of functional capacity in children, because they are similar to their habitual physical activity patterns in duration and intensity (Sargeant, 1989).

The following section will discuss the anaerobic characteristics of children such as muscle metabolism at rest and during exercise, muscle characteristics and changes with growth, and the oxygen uptake kinetics of children. The possible factors that influence the development of anaerobic performance in children will also be considered.

2.3.1.1 Muscle metabolism at rest and during exercise

2.3.1.1.1 Anaerobic metabolism

Muscle contraction requires energy (Eriksson, 1979). Energy is produced by chemically splitting adenosine triphosphate (ATP) into adenosine diphosphate (ADP) and phosphorous (P). The stored amount of ATP available for muscle contraction is limited and normal resting values are in the region of 4 – 5 mmol/kg of wet weight (w.w) muscle. Limited ATP supply within the muscle would be used within a few seconds so ATP is re-synthesised in the muscle. ATP re-synthesis can take place by three different means. Two are anaerobic (without oxygen) and one aerobic (with oxygen). The quickest method of obtaining ATP for muscle contraction is the anaerobic splitting of creatine phosphate (CP). The phosphate liberated from this splitting will form additional ATP as illustrated.

$$\mathbf{ADP + CP \rightarrow ATP + C}$$

Approximately 15 – 20 mmol/kg w.w of CP is stored in adult muscle and is immediately available for energy production (Karlsson, 1971a). The resyntheis of ATP from the splitting of CP and combining with ADP only allows energy production lasting approximately 5 - 10 seconds (Keul, 1972). Energy produced in this way is described as alactacid anaerobic capacity (Åstrand & Rodahl, 1986).

The second anaerobic process produces energy by splitting glucose or glycogen to pyruvate. Where there is a lack of oxygen, pyruvate is converted to lactate. Lactate has been related to fatigue during and following strenuous exercise and inhibits glycolytic enzymes and consequently glycolytic capacity (Pfitzinger & Freedson, 1997). One mol of glycogen will produce two mols of ATP. This reaction starts at the beginning of exercise and is responsible for the majority of energy production for the first 1 – 2 minutes of exercise. Energy produced in this way is described as the lactacid anaerobic capacity or anaerobic glycolysis. Anaerobic glycolysis is

limited by the accumulation of lactate in the muscle and by the subsequent acidosis that develops in the blood and the body. Anaerobic glycolysis obtains only two mol of ATP per mol of glycogen and is a relatively inefficient method of producing ATP.

Aerobic metabolism, the third method of ATP production, is achieved by oxididising pyruvate to carbon dioxide (CO_2) and water (H_2O), which yields a total of 38 mol ATP per mol of glycogen. The benefits of aerobic energy production are that more energy is produced per mol of glygogen with no accumulation of lactate. This process begins at the commencement of exercise and takes approximately 2 minutes to achieve the oxygen uptake to supply 100% of energy from aerobic metabolism. If the demand for energy exceeds the ability of the aerobic process to supply it, then anaerobic glycolysis will contribute to energy production. As a result, exercise time will be limited (Åstrand & Rodahl, 1986).

Studies of anaerobic metabolism of adults have been conducted since the early 1960s using a muscle biopsy technique (Bergstrom, 1962). The technique involves a section of muscle being removed from the intact muscle using a needle similar to an apple corer. The section of muscle is then snap frozen in liquid nitrogen for later analysis. Muscle biopsy has enabled a snapshot of *in vivo* muscle metabolism at rest, during exercise and post exercise and provided a greater understanding of muscle metabolism. Muscle biopsy is a technique well used in adult studies of muscle metabolism. Since the nature of muscle biopsy is very invasive, relatively few studies of muscle metabolism in children have been conducted due to the ethical and moral constraints of pediatric research (Rowland, 1996). Muscle biopsy studies of children have been generally limited to the work of Eriksson and his colleagues in the 1970s (Eriksson *et al.*, 1971; Eriksson *et al.*, 1973; Eriksson & Saltin, 1974; Eriksson, 1979), another three studies involving muscle biopsies on children were conducted in the 1980's (Fournier, 1982; Haralambie, 1982; Berg & Keul, 1988).

2.3.1.1.2 Muscle metabolism at rest

Precursors for energy metabolism, ATP, CP and glycogen as well as enzymes necessary to drive the reactions have been studied in the pediatric population using muscle biopsy. The first of these studies was conducted by Ericksson and his colleagues (Eriksson *et al.*, 1971). The authors took muscle biopsies from the vastus lateralis muscle of eight boys aged 13 years. Eriksson et al., (1973) reported that the resting concentration of ATP, CP and glycogen was 5.0, 17.0 and 69 mmol/kg w.w., respectively.

The paper by Eriksson et al., (1973) is one of the most frequently cited studies concerning the anaerobic potential of children. Ericksson and his colleagues (1973) conducted a training study with two groups of boys aged between 11 and 13 years of age. Resting concentrations of ATP were marginally lower (4.3 mmol/kg w.w.) than previously reported, but CP and glycogen concentrations were comparable with other studies conducted by Ericksson and his colleagues (1971;1974) (Eriksson *et al.*, 1973).

Ericksson et al. (1974) conducted a cross-sectional study of males aged between 11 and 16 years of age. The mean age of each of the groups was 11.6, 12.6, 13.5, and 15.5 years. The authors added the data from the 1971 study above (Eriksson *et al.*, 1971) and described the change in the concentration of CP and glycogen when younger participants were compared with their older peers. Resting concentrations of ATP were approximately 5 mmol/kg w.w. There was a tendency to higher resting CP concentrations with increasing age with values of 14.5 up to 23.6 mmol/kg w.w. in the older age groups. The same trend was observed in the resting concentration of glygogen with 54 mmol/kg w.w in the youngest age group. Resting glycogen concentrations of 70, 69 and 87 mmol/kg w.w were reported for the 12.6, 13.5, and 15.5 year old boys, respectively (Eriksson & Saltin, 1974). The authors compared their results with the resting concentrations in adults reported in Karlsson's (1971) study and Knuttgen and Saltin's (1972) study. ATP concentrations were found to be the same. Karlsson (1971) reported CP concentrations between 15 – 18 and 20 – 23 mmol/kg w.w., which were similar to those found by Ericksson and Saltin (1974). Resting glycogen concentrations were smaller than those reported by Karlsson (1971). Only the values of the oldest boys in Ericksson and Saltin's (1974) study were comparable with those of Karlsson (1971).

2.3.1.1.3 Metabolic enzymes

The activity of metabolic enzymes was also investigated in a few studies using muscle biopsy (Eriksson *et al.*, 1973; Haralambie, 1982; Berg & Keul, 1988). The majority of interest in this area surrounded the rate-limiting enzyme in glycolysis, phoshophructokinase (PFK) and the oxidative enzyme, succinate dehydrogenase (SDH).

Ericksson et al., (1973) were particularly interested in enzyme activity associated with energy production and examined resting activity of PFK and SDH in a group of thirteen boys aged between 11 and 13 years of age. Resting SDH activity was 5.4 μmol x (g x min) $^{-1}$. Ericksson et al., (1973) reported that this was approximately 20% greater than that observed by Gollnick et al.,

(1972). in sedentary adults PFK activity at rest was 8.4 μmol x (g x min) $^{-1}$, this level was less than 50% of that observed by Gollnick et al., (1972) in an adult population.

Berg and Keul (1988) investigated the biochemical differences in the muscle of three groups of children aged 4 - 8 years, 12-14 years and 16-18 years from muscle biopsy samples of vastus lateralis taken during surgical intervention. The authors observed that the concentrations of the enzymes linked to glycolytic metabolism (aldolase, pyruvate kinase, lactic dehydrogenase) were lower in the younger children (4 - 8 years) when compared with the older children (Berg & Keul, 1988).

In contrast to the findings of the previous papers (Eriksson *et al.*, 1973; Berg & Keul, 1988), Haralambie (1982) reported no differences in the concentrations of 10 glycolytic enzymes, including PFK, in muscle biopsy samples taken from the vastus lateralis of 7 girls aged between 13 - 15 years. Haralambie (1982) suggested that the possible reason for the different findings when compared with those of Eriksson et al., (1973) might have be due to the maturity differences of the two groups.

A training study conducted by Fournier et al., (1982) of 12 adolescent males aged between 16 and 17 years reported resting PFK activity of 28.1 μmol x (g x min) $^{-1}$ and SDH activity of 8.2 μmol x (g x min) $^{-1}$ The authors commented that these values were lower than those normally observed in adults (Fournier, 1982). The activity of both PFH and SDH were higher than that observed by Ericksson et al., (1974), but this may be due to the age of the participants in this study who would have been more mature than those of Ericksson et al., (1974).

Rowland (1996) suggests that an improved understanding of muscle metabolism in children may come from new noninvasive experimental techniques such as nuclear magnetic resonance spectroscopy (NMRS). NMRS technology enables the in situ examination of glycolytic activity in the muscle cell and can provide information on the changes in the concentration of inorganic phosphate (P_i), phosphocreatine (PCr) and pH (Rowland, 1996). Spreit (1995) questions the validity of NMRS, as the capacity of the chemical compounds in the skeletal muscle has not been calibrated. Calibration is achieved by assuming that the area under the ATP peaks of the NMRS spectra equate to the chemically determined ATP content. This relationship is used to quantify the area under the phospocreatine (PCr) peak. If the spectrometer does not see all of the intracellular ATP, then the estimated PCr will be too high. This has led to several investigators reporting resting PCr levels that are far higher than those determined by muscle biopsy (Spriet, 1995).

More recent studies using phosphorous 31 nuclear magnetic resonance spectroscopy (^{31}PNMRS) have suggested that the resting PCr and ATP concentrations of children are comparable with those of adults (Ferretti, 1994). Ferretti et al., (1994) reported resting ATP ($mM.kg^{-1}$) to be 13.6 arbitrary units (A.U.) in male and female children when compared with the resting ATP ($mM.kg^{-1}$) concentration of adults (11.9 A.U.).

2.3.1.1.4 Muscle metabolism during and after exercise

Muscle biopsy studies in the pediatric population have described the decrease in ATP, CP and glycogen during exercise and compared these results with those of adults (Eriksson *et al.*, 1971; Eriksson & Saltin, 1974). Other studies have examined the action of metabolic enzymes, PFK and SDH, before and after training (Eriksson *et al.*, 1973; Fournier, 1982).

Eight 13-year-old boys participated in maximal exercise on a cycle ergometer. Their ATP, CP, glycogen, blood and muscle lactate concentrations were measured prior to and post exercise (Eriksson *et al.*, 1971). The authors reported that the 13 year old boys ATP concentration decreased by 1.1 mmol/kg w.w., post exercise. The same trend was observed in CP, with the concentration decreasing by 12.1 mmol/kg w.w. Muscle glycogen decreased 35 mmol/kg w.w post exercise. Ericksson and his colleagues found the greatest individual variation in the post-exercise muscle and blood lactate concentrations. Mean muscle lactate concentrations at rest were 1.3 mmol/kg w.w. Mean post-exercise concentration was 11.3 mmol/kg w.w., but the range was from 6.3 to 14.6 mmol/kg w.w., which was lower than those reported by Karlsson (1971) in adult males (Ericksson et al., 1971). The authors were curious about the source of the large variation in post exercise lactate concentration and after observing that there was a variation in the maturational stage of the participants, correlated the post-exercise muscle lactate concentration with testicular size. The participants with the greatest testicular volumes had higher post-exercise muscle lactate concentrations. The authors concluded that the participants' level of sexual maturation exerted an influence on lactate production, but they were unsure of the factors that affected it (Ericksson et al., 1971).

Ericksson and Saltin (1974) reported similar findings of a lower post-exercise muscle and blood lactate concentration in their cross-sectional study of males aged between 11 and 16 years. The lowest concentrations were found in the younger (11.6 years old) when compared with the older group (15.5 years old). Lactate concentrations of the older group in the study were close to those reported by Karlsson (1971). Lower PFK values were suggested as the reason for attenuated muscle and blood lactate concentrations in the younger group when compared with the older

group. As a result, Ericksson and Saltin (1974) postulated that younger children might be handicapped in relation to providing energy for sports with a duration of 1 – 2 minutes.

In order to examine the impact of consistent structured physical activity on the muscle metabolism of children, training studies have been conducted (Ericksson et al., 1973; Fournier et al., 1982).

Ericksson and his colleagues conducted two different training studies in their 1973 paper. A total of thirteen males aged between 11 and 13 years participated in a four-month training program (series 1; n = 8) and a 6-week program (series 2; n = 5) designed to examine the impact of training on PFK, SDH and muscle cross-sectional area. Muscle biopsies were taken from vastus lateralis and for those participants in series 1 were analysed for glycogen, pyruvate, lactate, ATP, CP, glucose and glucose – 6 – phosphate concentrations. Series 2 biopsies were examined for PFK, SDH activity and changes to muscle cross-sectional area. Training for the series 1 study consisted of 34 sixty-minute sessions where the participants were engaged in a callisthenic warm-up, interval running and games. Series 2 training consisted of cycling for at least 20 minutes on the cycle ergometer (mean 29.8 min). Heart rates were recorded during each session. All participants recorded mean heart rates not less than 20 ($b.min^{-1}$) lower than their observed maximum heart rate during all training sessions.

The results of series 1 were a small increase in the resting concentrations of ATP and CP. There was a statistically significant difference in the resting concentration of muscle glycogen when before training values (53.9 mmol/kg w.w.) were compared with post training values (71.0 mmol/kg w.w.).Muscle glucose, glucose – 6 – phosphate and blood lactate showed similar values post training as pre-training. Series 2 participants showed significantly greater SDH and PFK activity post-training. PFK activity increased significantly after 2 and 6 weeks of training. The increase in SDH activity was only significant after 6 weeks of training. There was no difference in the percentage of slow twitch fibres examined after 2 and 6 weeks of training. No pre-training samples were reported. Ericksson and his colleagues concluded that training increased the glycolytic potential of the participants, this lead to greater glycogen depletion post-exercise when compared to pre-training values. The adaptation to training was similar to that observed in adults (Ericksson et al., 1974).

Twelve 16 – 17 year old males participated in training four times per week for three months. The participants were divided into an endurance-training group and a sprint-training group. The endurance (END) group participated in progressively increased runs from two repetitions of 10 minutes to two repetitions of 30 minutes. Intensity ranged from 60 – 70% maximal heart rate to

80 – 90% maximal heart rate towards the end of the study. The sprint (SP) group participated in interval runs from 50 – 250 metres and occasionally ran up stairs. Muscle biopsies were taken from vastus lateralis and analysed for changes in muscle fibre type and changes in SDH and PFK activity as a result of training (Fournier et al., 1982).

The END group exhibited a significant increase in slow twitch, fast twitch a and fast twitch c cross sectional area, but no changes in % fibre type distribution as a result of training. They also showed a significant increase in SDH activity post-training. The SP group did not show any significant increases in muscle cross-sectional area or % fibre type distribution. PFK activity significantly increased as a result of training. The authors concluded that END training adequately stimulated a response in muscle fibre and oxidative (SDH) enzymes that was similar to that seen in adults. The failure of SP training to adequately stimulate muscle fibres and a lower PFK activity in the SP group differed from findings in adults (Fournier et al., 1982).

2.3.1.1.5 Section summary

The following table provides a summary of muscle metabolism at rest and post-exercise from early muscle biopsy studies on children when compared to adults.

Table 2.6 Substrate availability and utilisation in muscle of pre-adolescent boys as compared with adolescents and young adults (Adapted from Bar-Or, 1983)

	Resting Values		
Substrate	Concentration in muscle (mmol/kg Wet Weight)	Compared with Older Individuals	Utilization rate during exercise
ATP	3.5 – 5	No change with age	Same as adults
CP	12 – 22	Lower in children	Same or less than adults
Glycogen	45 – 75	Lower in children	Much less than adults

Based on (Karlsson, 1971b; Eriksson & Saltin, 1974; Eriksson, 1979)

2.3.1.1.6 ^{31}PNMR studies of metabolic changes in the muscle during exercise in children

As discussed previously early studies of pediatric muscle metabolism were conducted using muscle biopsy. These studies were extremely invasive and consequently none have been conducted since the 1980's. The following studies have used a non-invasive ^{31}PNMRS technology and have compared children and adults participating in the same study.

Gastrocnemius muscle phosphate (P_i,) phosphocreatine (PCr) and pH were measured at rest, during progressive maximal treadle exercise and recovery in 10 pre-pubertal children and 8 adult participants. Resting values of pH, P_i/PCr and PCr/ATP in the pre-pubertal group were not significantly different from those of the adult group. The post exercise results indicated that the adults performed significantly greater work than the children did. The adults also showed a significantly decreased P_i/PCr ratio and a significantly lower pH when compared to the pre-pubertal group (Zanconato *et al.*, 1993).

Zanconato and her colleagues identified two phases or slopes of P_i/PCr increase and pH decrease during exercise in both of the groups. The first phase, which was also termed the fast phase, was connected to the low-intensity section of the progressive exercise test. There were no significant differences in the P_i/PCr increases during this phase when the adults were compared to the children. The transition from the fast phase to the slow (second) phase was visually inspected in six adults and five children. The transition occurred at approximately 40% of the maximal work rate in the adults and approximately 60% of the maximal work rate in the children (Zanconato et al., 1993). The slow phase of the exercise test, which is related to higher intensity exercise, indicated that the adults had significantly greater ratios of P_i/PCr when compared to the children. Increases in the ratios of P_i/PCr are indicative of accelerated anaerobic glycolysis. The decrease in pH concentration showed the same general trends as the P_i/PCr ratio, with the adults experiencing significantly greater acidosis when compared to the children (Zanconato et al., 1993).

In a discussion of the possible mechanisms accounting for the differences observed in the slow phase of the exercise test, the authors suggested that the similar response in the fast phase of exercise might indicate that the children have a similar rate of oxidative metabolism when compared to the adults. The different responses observed in the slow phase may suggest a growth-related difference in high-intensity exercise metabolism. Zanconato et al., (1993) postulated that there might be two reasons for this difference in high-intensity exercise metabolism. The first concerned a possible higher rate of muscle oxidative phosphorylation during heavy exercise in children. A higher rate of oxidative phosphorylation could result from factors such as a greater delivery of oxygen from the capillary blood, the delivery of substrates or a greater mitochondrial density. All of these factors would account for a greater oxygen dependent ATP generation during high-intensity exercise (Zanconato et al., 1993). However, a more efficient oxidative mechanism should not inhibit the glycolytic capability. At some point, as the intensity of exercise increases, glycolysis and the resultant lactate production would be

necessary to regenerate ATP (Zanconato et al., 1993). An alternative postulation, that the functional glycolytic capability of children may not be sufficient to meet the energy requirements of high-intensity exercise, and that as a result children may reach muscular fatigue earlier, was put forward. This point is supported by the finding that the children achieved a Pi/PCr ratio that was only 27% of that of the adults (Zanconato *et al.*, 1993).

Kuno et al. (1995) has also examined the muscle metabolism of children using ^{31}PNMRS. The authors investigated the changes in the values of PCr (PCr + Pi) and pH in males from 12 - 15 years of age when compared with those of adults after maximal treadle exercise. Higher values of PCr and pH were found in the children when compared to the adults after the exhausting exercise bout (Kuno *et al.*, 1995).

Table 2.7 presents a summary of the literature to date using ^{31}PNMRS to investigate the muscle metabolism of children after exercise.

Table 2.7 Muscle metabolism during exercise from 31 PNMRS studies (Adapted from Van Praagh, 2000)

Author	Methods	Age (yrs)	Sex	PCr (PCr + Pi)	Intracellular pH
(Zanconato *et al.*, 1993)	^{31}PNMRS (gastrocnemius muscle)	7-10	male	Higher*	Higher*
(Kuno *et al.*, 1995)	^{31}PNMRS (quadriceps muscle)	12-15	male	Higher*	Higher*

*Higher than adults in the same study.

2.3.1.1.7 Section summary

Resting values of muscle ATP and PCr have been shown to be similar to values observed for adults. The ability, however, for children to exhibit the same glycolytic capacity as adults appears to be limited by smaller resting concentrations of glycolytic enzymes, although the data is somewhat limited. More recent ^{31}PNMRS studies have observed that the values of PCr and pH in children and adolescents are higher than those of adults after exhausting exercise.

2.3.1.2 Changes in blood metabolites after exercise

2.3.1.2.1 Blood lactate

Post-exercise blood lactate is an indicator of glycolytic metabolism during exercise (Pfitzinger & Freedson, 1997). Since the earliest studies of muscle metabolism, blood and muscle lactate concentrations after exercise have been reported as being smaller in children than in adults (Åstrand, 1952; Eriksson *et al.*, 1971; Eriksson & Saltin, 1974). Since these early studies, a number of studies have been undertaken that have examined post-exercise lactate concentrations in children (Fellman *et al.*, 1988; Mero, 1988; Falgairette *et al.*, 1990; Williams & Armstrong, 1991; Falgairette *et al.*, 1993; Welsman *et al.*, 1994; Pianosi *et al.*, 1995; Hebestreit *et al.*, 1996; Armstrong *et al.*, 1997). In a discussion of studies investigating post exercise lactate concentrations, a number of factors must be taken into account when comparing the results of these studies. The first of these is that post-exercise lactate concentrations reflect all the process in which lactate is produced and removed (Stanley, 1985). Consequently, post-exercise lactate concentrations provide an indication of the degree to which the exercise had placed stress on the metabolic system, but is not an exact measure (Van Praagh, 2002). Other considerations such as: exercise mode, exercise protocols (Wingate vs peak oxygen uptake), sampling times and participant motivation may also affect post-exercise lactate concentrations (Pfitzinger & Freedson, 1997). Specific exercise protocols, depending on their length, will stimulate the glycolytic system to different degrees. For example, muscle metabolism during a Wingate Anaerobic Test lasting 30 seconds will stimulate the ATP-PC and alactic systems to a greater degree than the lactacid system. In contrast, a peak oxygen uptake test lasting 12 minutes will stimulate all systems, the aerobic system would predominate, the lactacid system would come into play towards the end of the test (Pfitzinger & Freedson, 1997). Mero (1988) illustrated this when he investigated post-exercise lactate concentration in 25 pre-pubertal males (n = 19 trained; n = 6 controls) who completed a modified 15-second Wingate test, a modified 60-second Wingate test and a peak oxygen uptake test (mean time 23 minutes). Higher post-exercise lactates were observed after the 60 second Wingate when compared to the peak oxygen uptake (60.6%) and the 15 second Wingate test (68.7%) (Mero, 1988). In an investigation of post-exercise lactate in 10 – 12 year old Bolivian boys found that post-exercise lactate concentrations were similar after a peak oxygen uptake test and a 30 second Wingate test (Fellman, 1994). The findings of Mero (1988) and Fellman et al., (1994) support the contention that the 30 second Wingate Test may not be sufficient to maximally stress the glycolytic system in children (Pfitzinger & Freedson, 1997).

Ericksson et al. (1973) found a correlation between testicular size and post-exercise lactate concentration. Later studies suggested that there was a strong relationship between circulating testosterone and post-exercise lactate concentrations (Inbar & Bar-Or, 1986; Fellman *et al.*, 1988; Mero, 1988; Falgairette *et al.*, 1990). Inbar and Bar-Or (1986) suggested that the concentration of

circulating testosterone might be related to the glycolytic capacity of children as had also been found in rats (Krotkiewski, 1980). Other studies, however, have found no correlation between testosterone concentration and post-exercise lactate concentration (Welsman *et al.*, 1994; Armstrong *et al.*, 1997).

As illustrated in Table 2.8 below, post-exercise lactate concentration shows an upward trend as children grow and mature. The reason for the smaller post-exercise lactate concentration is commonly thought to be related to the lower PFK activity in the muscle identified by Ericksson et al., (1973).

Table 2.8 Peak blood lactate concentration (Adapted from Pfitzinger and Freedson, 1997)

Study	N	Age	Peak blood lactate concentration m/mol	Findings
Ericksson and Saltin (1974)	33	11.6 12.6 13.5 15.5	7.9 ± 0.5 9.6 ± 0.6 9.2 ± 0.3 10.5 ± 0.9	Higher lactates in older participants
Cumming et al., (1980)	309	4 – 5 6 – 7 8 – 9 10 – 12 13 – 15 16 - 20	(m) 9.5 (f) 10.4 (m) 9.1 (f) 9.5 (m) 9.9 (f) 10.2 (m) 10.2 (f) 10.2 (m) 11.3 (f) 11.6 (m) 12.1 (f) 10.4	Males exhibited higher lactates with age. Same trend not observed in females. Higher lactates at age 4 – 5. Not ages 13 – 20.
Paterson et al., (1987)	18	11 12 13 14 15	7.0 ± 1.4 7.7 ± 1.7 9.0 ± 1.9 9.0 ± 1.7 10.1 ± 1.5	Peak lactate concentration increased with age, authors cautioned that older subjects more highly trained
Williams and Armstrong (1991)	191	Tanner 1 2 3 4 5	(m) 5.3 (f) N/A (m) 5.0 (f) 5.8 (m) 4.7 (f) 5.7 (m) 5.8 (f) 6.6 (m) 5.8 (f) 5.7	Higher lactate in girls. Lactate concentration not related to age or Tanner stage

Although, as previously discussed, lower activity of glycolytic enzymes has been found in children, other studies have suggested that a lower glycogen concentration and utilization in the muscle (Inbar & Bar-Or, 1986; Zwiren, 1989) and a reduced sympathetic drive (Berg & Keul, 1988) may also be reasons for smaller lactate concentrations in children. Pfitzinger and Freedson (1997) suggest that as the literature on post-exercise lactate concentration increases, the understanding of the mechanisms of the smaller concentrations in younger, when compared with older children may become apparent. Until that time, an understanding of the mechanisms of smaller concentrations remains problematic (Rowland, 1996).

2.3.1.2.2 Acid/Base balance

Limited studies of the post-exercise acid/base balance (pH) of children have been conducted. Post-exercise acid/base balance has been reported in pH (units) or as base excess (Inbar and Bar-Or, 1986). Maximal base excess was investigated in 172 students aged between 11 and 15 years who had undergone a peak oxygen uptake test (Gaisl, 1977). The authors reported that maximal base excess increased with age (Gaisl and Buchberger, 1977). The pH response to maximal exercise in males (n = 103) aged 12 to 15 years was investigated in a study conducted over four years (Matejkova et al., 1980). An age-related decrease in the pH response to maximal exercise was observed in this study and the authors suggested that the maximal level of post-exercise acidosis was related to age (Matejkova et al., 1980). In a more recent study, Hebestreit et al., (1996) found that plasma hydrogen ion concentration increased more in adults when compared with boys aged between 8 and 11 years who had undergone 30 seconds of high intensity cycle exercise. These findings support the suggestion that boys have a smaller glycolytic capacity when compared with adults. Hebestreit and his colleagues (1996) suggested that the age-related difference in blood acidosis reported their study were in agreement with the findings of Zanconato et al., (1993) of a higher muscle pH in children when compared to adults after maximal treadle exercise.

Recently, Ratel et al., (2002) investigated the acid/base balance of 11 boys (mean age 9.6 ± 0.7) after repeated cycling sprints and compared the findings with those of 10 adults (mean age 20 ± 0.8). The authors chose ten 10's sprints with a 30 second recovery to maximally stress the lactic acid system of the subjects. The boys exhibited a decrease in pH of 0.07 units from rest to the sixth cycle sprint. pH remained unchanged until after the tenth sprint. In contrast the adults' pH decreased 0.23 units from rest to the tenth sprint. The adults exhibited a significant decrease in pH from the boys after the second sprint ($p < 0.05$), this trend continued until the adults exhibited a 1.5 fold lower pH ($p < 0.001$) after the final sprint (Ratel et al., 2002). Ratel and his colleagues

(2002) suggested that the smaller pH response of the boys when compared to the adults was still to be elucidated, but research suggested that children were better equipped for aerobic than anaerobic exercise.

2.3.1.3 Muscle characteristics and changes with growth

2.3.1.3.1 Muscle mass

The percentage of muscle mass that contributes to physical activity in 5 year-old males is equivalent to approximately 42% of their body mass. This increases to approximately 54% of body mass by the time the male child has reached 18 years of age. This equates to muscle weight of 7.5 kg to 37 kg, which is approaching a five-fold increase from childhood (Malina, 1969). When boys have reached 6 to 7 years of age the boys have been shown to have a larger absolute and relative (kilogram of muscle per kilogram of body mass) when compared with that of girls (Malina, 1969). Ferretti et al., (1994) suggested that the patterns of muscle development exhibited by boys may account for the large gender differences associated with strength during childhood and adolescence.

2.3.1.3.2 Hormonal effects on muscle mass

The changes in muscle mass that are observed in children as they progress through puberty have been largely attributed to the influence of hormones (Preece, 1986). The influence of testosterone on the muscle mass of males is of particular interest. Testosterone is suggested as one of the most active contributors to the anabolic processes in the muscles. Other hormones such as growth hormone, thyroid hormones, somatomedians and insulin are also known to be important contributors to the development of muscle mass in children (Florinin, 1987). Testosterone, in male children, increases approximately four times during early puberty. Testosterone levels will increase by another twenty times by the time the child reaches late puberty (Blimkie, 1998).

2.3.1.3.3 Fibre differentiation

In relation to development changes in muscle fibre type, Colling-Saltin (1980) observed that the proportion of type II fibres in early childhood was lower than that of adults. The distribution of type II fibres has been seen to reach adult proportions during late adolescence (Hedberg, 1976; Fournier, 1982). In contrast, (Bell *et al.*, 1980) demonstrated that the histochemical profile of the muscle fibres of 6 year-old children was similar to that of young adults.

Type II muscle fibres possess characteristics that make them more adapted for anaerobic performance. They have high concentrations of glycolytic enzymes and greater contractile speeds

than type I fibres (Åstrand *et al.*, 1986). The increase in the proportion of type II muscle fibres that is supported by some of the limited research into the muscle fibre types of children would impact on the ability of the child to perform anaerobically.

2.3.1.4 Oxygen uptake kinetics

Oxygen uptake kinetics describes the mechanics of oxygen uptake during exercise. The oxygen uptake kinetics of children when compared with that of adults has been observed to be both qualitatively and quantitatively different (Armon et al. 1991). Armon et al. (1991) support this notion with observations from a study investigating the oxygen uptake dynamics during high-intensity exercise in children and adults. From a qualitative perspective, Armon et al. (1991) observed that fewer children developed oxygen drift at high-intensity exercise when compared with the adults in the study. In those children that developed an oxygen drift, it was smaller in magnitude (absolute terms) when normalised for body size when compared with the oxygen drift of the adult sample (Armon *et al.*, 1991). Quantitatively, the oxygen cost of high-intensity exercise ($\dot{V}O_2$ per watt) is higher in children, despite the oxygen drift that was observed in some of the children (Armon et al., 1991). The children in this study (1991) were also able to respond more quickly to the increased demands for oxygen at high-intensities when compared with the adults. This would lead to a greater oxygen cost of high intensity exercise in children when compared to adults. Armon et al. (1991) questioned whether the higher oxygen cost of exercise (per watt) in children was indicative of a more effective cardio respiratory response in children, or alternatively, or whether it was representative of a lower ability to generate ATP through anaerobic processes. Lower lactate levels observed in immature children when compared with their more mature counterparts supports the latter suggestion (Eriksson *et al.*, 1971; Eriksson & Saltin, 1974; Cumming *et al.*, 1980; Cumming, 1985). Armon et al. (1991) reinforced that the measurement of blood lactate concentration in children may not be indicative of actual lactate production because of the possible imbalance between lactate generation and removal. It has been suggested that children may clear lactate more quickly than adults due to an increased sympathetic drive (Zwiren, 1989).

2.3.2 Anaerobic performances of children

2.3.2.1 Short term anaerobic performance in children

The majority of studies investigating anaerobic performance in children have focused on short-term (6-30 second) cycle ergometer tests (Inbar & Bar-Or, 1986; Blimkie *et al.*, 1988;

Vandewalle *et al.*, 1989; Falgairette *et al.*, 1990; Mercier *et al.*, 1992; Falgairette *et al.*, 1993; Carlson & Naughton, 1994; Hebestreit *et al.*, 1996; Armstrong et al., 2001; Doré et al., 2001; Duché et al., 2002; Ratel et al., 2002; Doré et al., 2003; Martin et al., 2003; Martin et al., 2004). Two different types of tests have been employed: the Force-Velocity test and the Wingate Anaerobic Test (WanT). Both the Force-Velocity Test and the WAnT have been described as tests of anaerobic power. The Force-Velocity test involves a series of short all-out tests on a cycle ergometer against increasing braking forces. Maximal power recordings are made during the test, which usually lasts between 5 and 10 seconds. The breaking force continues to increase until there is no increase in the power generated by the participant. This series of tests generates a power-velocity curve (Vandewalle *et al.*, 1989; Falgairette *et al.*, 1990; Mercier *et al.*, 1992; Falgairette *et al.*, 1993). The WAnT is a 30-second all-out test conducted on a cycle ergometer against constant braking forces applied according to the participant's mass. The WAnT provides information about the participant's peak and mean power (watts) and relative peak and mean power ($watts.kg^{-1}$) (Inbar & Bar-Or, 1986; Blimkie *et al.*, 1988; Carlson & Naughton, 1994; Hebestreit *et al.*, 1996).

Researchers investigating the anaerobic performances of children using the Force-Velocity Test and the WAnT have observed an age-related increase in peak and mean power in cross-sectional studies (Inbar & Bar-Or, 1986; Blimkie *et al.*, 1988; Vandewalle *et al.*, 1989; Mercier *et al.*, 1992; Carlson & Naughton, 1994). Limited longitudinal studies investigating anaerobic performance using Force-Velocity Tests (Duché et al., 1992; Martin et al., 2004) and WAnT (Armstrong et al., 2001) have confirmed that peak and mean power increase with increasing age of the participant (Duche, 1992; Martin et al., 2004). Falk (1993) examined the anaerobic performances of a sample of participants that were classified as pre-pubertal (n = 16), mid-pubertal (n = 15) and post-pubertal (n =5) in a mixed cross-sectional longitudinal study. Their findings (1993) confirmed those of Duché et al., (1992). The following figures illustrate an age related increase in peak power (Figure 2.5) and mean power (Figure 2.6) from Wingate Anaerobic tests.

Figure 2.3 Age- related increase in peak power from Wingate Anaerobic Test (Adapted from Bar-Or, 1983)

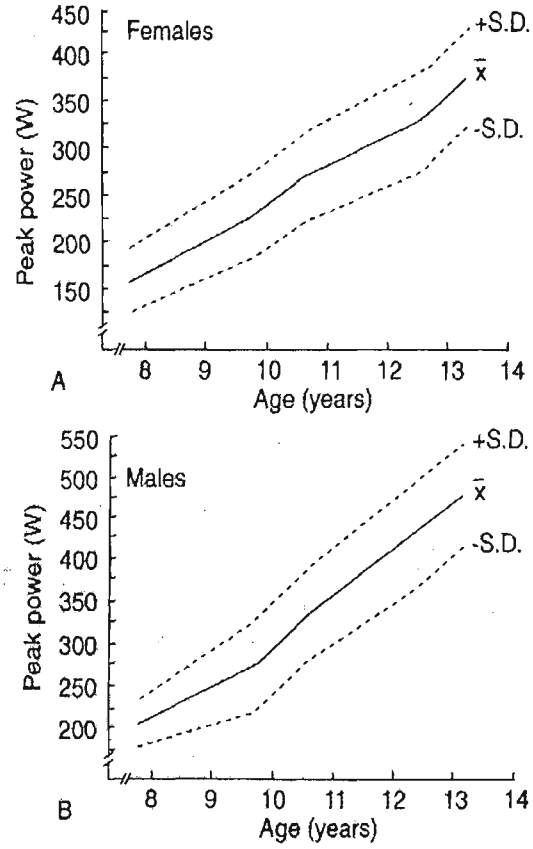
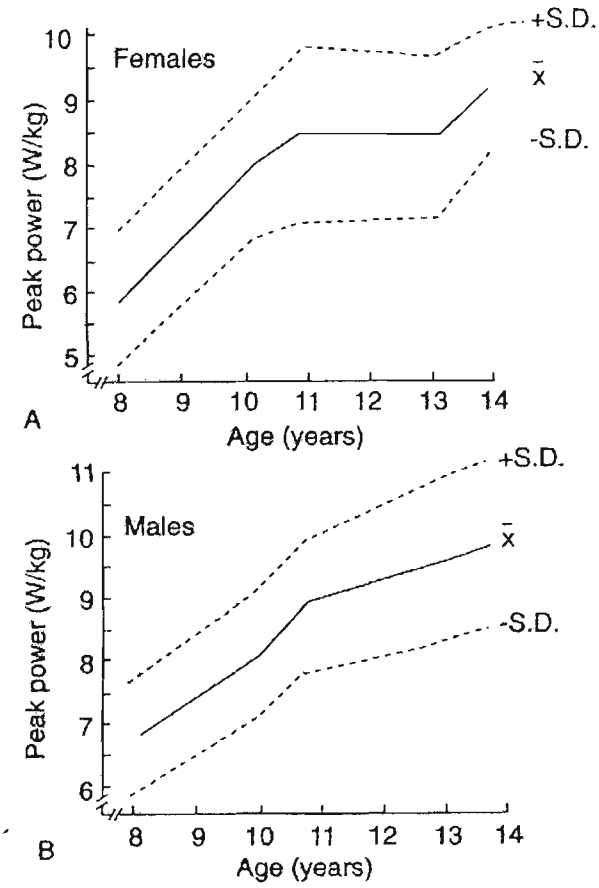


Figure 2.4 Age related increase in mean power (watts/kg) from Wingate Anaerobic Tests (Adapted from Bar-Or, 1983)



2.3.2.2 Anaerobic performances to exhaustion in children

Limited numbers of studies examining the anaerobic performance of children using AOD have been reported in the literature (Carlson and Naughton, 1993; Buttifant et al., 1996; Naughton et al., 1997). Table 2.9 presents the findings of accumulated oxygen deficit in children to date.

As mentioned previously, studies comparing the AOD must be viewed with caution due to the different methodologies to determine the AOD employed by different researchers (Bangsbo, 1996a; Gastin, 1994). The studies above have all used the same methodology in the same laboratory, so it is hoped that they provide a reasonable comparison.

The studies above have found a larger AOD with treadmill exercise (Buttifant et al. 1996) when compared to cycle ergometer exercise (Carlson and Naughton, 1993) in pre-pubertal males. The AOD is greater in pubertal participants (Naughton et al. 1997) than in pre-pubertal participants

(Buttifant et al. 1996; Carlson and Naughton, 1993). There are no differences between the performances of the pre-pubertal males and females (Carlson and Naughton, 1993), but large differences in the AOD were present when the male and female pubertal participants were compared in the study conducted by Naughton et al. (1997).

Table 2.9 AOD in the pediatric population

Study	Participants	n	Exercise Mode	Exercise Intensity	AOD (ΣL)	AOD ($\Sigma mL.kg^{-1}$)
Carlson & Naughton (1993)	Pre-pubertal males	9	Bicycle ergometer	110%	1.3	35.3
		9		130%	1.3	37.1
		9		150%	1.3	36.8
Buttifant et al. 1996	Pre-pubertal non-asthmatic males	10	Treadmill	110%	1.7	51.5
		10		130%	1.6	47.0
Naughton et al. 1997	Pubertal males	8	Treadmill	120%	4.3	71.5
		8		130%	4.1	67.6

The studies examining the anaerobic performance of children using the AOD have reflected those findings of an increase in anaerobic performance measured by the AOD with increasing participant age.

2.3.2.2.1 The issues of the use of accumulated oxygen deficit with children

Two recent reviews have discussed the issues surrounding the use of the AOD as a means of measuring anaerobic performance in children (Carlson & Naughton, 1998). Carlson and Naughton (1998) and Naughton and Carlson (1998) suggested that the controversies surrounding the AOD with the adult population were likely to become more complex with the pediatric population.

The first of the issues is related to the range of submaximal intensities available to determine the relationship between submaximal oxygen uptake and exercise intensity (submaximal economy). At low exercise intensities there is likely to be an increased oxygen cost of exercise, especially on the treadmill, due to extra energy required to coordinate and balance the child. At high exercise intensities there is a need to avoid exercise intensities that incur an oxygen drift that will prevent

the attainment of steady state. Children may, however, be able to maintain a higher exercise intensity without oxygen drift when compared to adults (Armon *et al.*, 1991). Achieving the greatest number of submaximal economy relationships is perhaps more difficult in children because of their smaller work rate range (Naughton and Carlson, 1998).

The second issue concerns time-efficient test procedures. The most time-efficient testing procedures desirable with pediatric populations are not possible if the methodology proposed by Medbø et al., (1988) of at least 10 submaximal exercise bouts is to be employed with pediatric populations (Naughton and Carlson, 1998).

The third issue relates to some evidence suggesting that children move quickly from anaerobic to aerobic provision of energy for exercise (Naughton and Carlson, 1998). Factors such as higher susceptibility to hypoxic conditions and a smaller capacity for carbon dioxide storage (Poage, 1987; Springer, 1988) may have led to adaptations in the child's oxygen uptake kinetics that mask the capacity for anaerobic power at the onset of exercise (Naughton and Carlson, 1998). The possibility of an increased sensitivity to hypoxia in children may affect the validity of the assumption that a constant efficiency between submaximal and supramaximal exercise exists. This may lead to an underestimation of the predicted oxygen demand in children (Naughton and Carlson, 1998).

In light of these issues, Carlson and Naughton (1998) suggested that further investigations into the use of the AOD with pediatric populations be conducted. Specific areas of investigation included:

- Determining the relationship between different exercise modes and AOD.
- Detecting training adaptations with different pediatric populations using AOD.
- Whether different submaximal protocols resulted in different linear prediction equations.
- What is the developmental profile of AOD?

Investigations such as those indicated above may address some of the issues associated with the use of AOD to measure anaerobic performance in children and may make it a more attractive means of measuring anaerobic performance in children (Carlson and Naughton, 1998).

Chapter 3

METHODOLOGY

3.1 Introduction

This chapter describes the participants and the general data collection procedures common to more than one study in this thesis. Procedures that relate to specific study aims are presented in the relevant chapters.

3.2 Description of participants

Pre-pubertal, pubertal and post-pubertal populations were targeted for the research studies described in this thesis. Data were collected on a total of 60 male participants. Their ages ranged from 8 to 17 years. The participants who volunteered for these studies were students of local primary and secondary schools or members of sporting clubs.

All volunteers attended formal schooling; consequently they regularly participated in physical activity that was part of the school curriculum. In addition, all of the participants participated in sporting activities outside the school environment, which equated to at least 2 hours of additional physical activity per week. The range of additional physical activity was approximately 2 – 8 hours. Pre-pubertal participants generally participated in less additional physical activity when compared with post-pubertal participants. The range of descriptive characteristics of the participants is presented in Table 3.1.

Table 3.1 Descriptive characteristics of the participants

Study	N	Age (yr)	Mass (kg)	Height (cm)	Peak oxygen uptake ($L.min^{-1}$)	Peak oxygen uptake ($mL.kg^{-1}.min^{-1}$)
One	30	8 - 17	35 - 72	143 - 177	2.12 - 4.99	59 - 69
Two A	10	9 - 12	30 - 53	146 - 159	0.99 – 2.16	52 - 70
Two B	10	8 - 12	30 - 51	135 - 161	-	-
Three	10	8 - 13	30 - 51	135 - 166	1.69 – 3.66	55 - 75

3.3 General data collection and procedures

Prior to the commencement of each study, the Victoria University Human Ethics Research Committee approved the procedures. Study participants and their parents/guardians were informed of the purpose, procedures and the risks of the studies in both written (Appendix 1) and verbal assent before informed consent was given by the parents/guardian and the participants.

In accordance with the guidelines of the Victoria University Human Ethics Research Committee, participants were informed that their participation in the study was voluntary and that they were free to withdraw from the study at any time without prejudice.

3.4 Assessing Sexual Maturation

Specifically, in Study One the Tanner Scale (Tanner, 1962) and the concentration of testosterone in saliva were used as objective markers of sexual maturity. A comprehensive explanation of the methodology used to establish the concentration of testosterone in saliva is addressed in Chapter 4. In studies Two and Three, the Tanner Scale was used as the only objective marker of sexual maturity.

3.4.1 The Tanner Scale

Tanner (1962) developed a pictorial scale, which represents the five stages of sexual maturity. The Tanner Scale uses the changes in the secondary sexual characteristics such as the growth of pubic hair and the enlargement of the testes to differentiate between children at different developmental stages. In the pictorial representation of the scale, T1 represents immature sexual characteristics and T5 represents mature sexual characteristics (See Appendix 2).

For the purposes of these studies, parents of participants aged between 8-12 years were asked to identify their child's stage of sexual maturity. In the first instance parents were informed of the use of the Tanner Scale as an indicator of sexual maturity in the Parent's Letter (Appendix 1.5). Once the study commenced, the parents were sent a letter alerting them to the fact that the Tanner Scale pictorial representations would be forwarded to them in the mail in the following week. They were also encouraged to contact the investigator if there were any queries. Participants who were aged between 13 and 17 years were asked to identify their own stage of sexual maturity in private. A pictorial representation of the Tanner Scale was placed in a manila folder along with the participant's record sheet. The participants were briefed prior to their viewing of the Tanner Scales. The participant was asked to view the Tanner Scale pictorials and rate themselves against them. At all times an emphasis was placed on the confidentiality of the ratings and on the importance of being as accurate as possible. A reliability of 89.9% has been reported in the self-reporting of Tanner Scale indices by males (Matsudo and Matsudo, 1993).

3.4.2 Laboratory procedures

3.4.2.1 Laboratory familiarisation

A laboratory familiarisation session was conducted for all participants prior to the commencement of testing. Specific equipment and procedures were introduced to the participants in a non-threatening environment. During the familiarisation session, the participants were instructed in treadmill running and introduced to the wearing of the Polar™ heart rate monitor and breathing through the respiratory valves.

3.4.2.1.1 Treadmill familiarisation

Instruction in treadmill running involved a demonstration, by the researcher, of the method of mounting and dismounting the moving treadmill. The participants were given the opportunity to practise these skills with the researcher positioned behind the participant to ensure his safety. The participant was then given the opportunity to walk and run on the treadmill at various speeds. While the participants were undergoing their familiarisation, technique points were given in areas such as where to focus their line of sight (i.e. not looking down at the treadmill). The participants were encouraged to aim to attain their normal gait pattern as soon as possible. In order to facilitate this, encouragement and feedback was given to the participants, including phrases such as "Just imagine that you are walking down the street" or "Just imagine that you are running in the park".

The time taken to complete the familiarisation with the treadmill varied according to the participants' prior experience. Each of the participants was given at least 10 minutes to familiarise himself with the equipment. If the participant was not comfortable, he was given the opportunity to continue his familiarisation until he was satisfied.

3.4.2.1.2 Heart rate monitor familiarisation

Polar ™ Heart Rate monitors were used to record the heart rate of the participants throughout all testing. Familiarisation with the heart rate monitor included an explanation, in simple terms, of how the heart rate monitor worked. The participant was then fitted with the heart rate monitor. He was given the opportunity to observe his heart rate while resting and during exercise.

The Polar™ heart rate monitor consists of an elasticised chest strap with in-built electrodes. Before the chest strap was placed on the child, it was checked for fit and comfort. Once the correct fit had been made, the electrodes were moistened with water and the chest strap secured

around the child. The Polar™ heart rate monitor uses radio telemetry to transmit the heart rate signal picked up by the chest electrodes and then displays the heart rate on the watch receiver.

3.4.2.1.3 Respiratory valve familiarisation

Two-way Hans Rudolf respiratory valves were used throughout the testing. Mouthpieces of various sizes were available to facilitate a comfortable fit into the mouth of even the smallest participants. The familiarisation with the respiratory valve included the selection of the appropriate mouthpiece for each participant. Once the mouthpiece was selected, it was attached to the valve and the participant was encouraged to breathe through the valve. Smaller children used smaller mouthpieces with the concomitant respiratory valve which was smaller than those used for adults. While the participant was breathing through the valve, he was instructed in the appropriate communication signals to be used while the mouthpiece was in the mouth thereby preventing verbal communication.

After the mouthpiece familiarisation was complete, the mouthpiece was attached to the headset. The headset device consisted of an adjustable head circumference band and adjustable arms that extend from the headband down to the mouthpiece. Appropriate adjustments to the circumference band and the attachment arms were made to ensure a secure and comfortable fit.

3.4.2.1.4 Information provided during the familiarisation session

Procedural details and specific demands of the testing were discussed with all potential participants during the familiarisation session. The participants were also reminded of their ability to withdraw from the testing at any time without prejudice.

3.4.3 The ergometers used during data collection

3.4.3.1 Treadmill

A Quinton™ (24-72) motorised treadmill was used for Studies One and Two. Study Three used a Quinton ™ Q65 (Series 90) treadmill. The treadmill was calibrated prior to every submaximal and supramaximal test.

3.4.3.2 Cycle ergometer

A CYBEX™ (met, 100 Lumex) electronically –braked cycle ergometer, which could be adapted for constant power or isokinetic resistances, was used in studies Two and Three. The CYBEX™ cycle ergometer provided screen displays of time, power and pedal frequency throughout all

testing. Toe stirrups were used at all times during testing. The children were also requested to remain seated on the cycle ergometer during testing.

3.4.4 Procedures for the collection of metabolic data

The peak oxygen uptake, respiratory exchange ratio (RER) and submaximal oxygen uptake were the main metabolic data collected and utilised during the studies. The metabolic data was determined using open circuit analysis of expired air. Room air was inhaled through a two-way Hans Rudolf respiratory valve. Expired air was exhaled into a mixing chamber through a lightweight tube with a diameter of 5 cm. A Pneumoscan ventilometer with a Mark 2 turbine flow transducer was connected to the mixing chamber. The estimated accuracy of the Pneumoscan ventilometer was ± 3%. The ventilometer was calibrated before and following each test with a 3-litre calibration syringe (Medical Graphics Corporation). Calibration of the ventilometer was flow-rate independant.

Applied electrochemistry analysers (S-3A (O_2) and CD-3A (CO_2) determined the fractions of O_2 and CO_2 of the gas samples pumped from the mixing chamber to the analysers at a rate of 300ml min^{-1}. Alpha gas samples (CIG, Melbourne) were used to calibrate the oxygen and carbon dioxide analysers before and immediately after each test. A-D converters connected the metabolic equipment to an on-line PC that calculated data at 15-second intervals. (An example of calculated metabolic data is presented in Appendix 3).

3.4.5 Protocols adapted for the determination of peak oxygen uptake

All of the studies used the same method of determining the peak oxygen uptake on a motorised treadmill. However, the treadmill used in Studies One and Two (Quinton™ (24-72) was different to that used in Study Three (Quinton™ Q65 (Series 90)). The peak oxygen uptake was determined using an incremental exercise protocol with a commencing treadmill speed of 6 kilometers per hour (km. hr^{-1}) at a constant grade of 6%. Increments of 1 (km. hr^{-1}) were applied every minute until the participant reached volitional exhaustion.

To determine whether a peak oxygen uptake had been achieved the following criteria were applied: a heart rate greater than 95% of the predicted maximal heart rate; a RER greater than 1.0; or a leveling off of oxygen uptake with increasing workload (Zwiren 1989). The final criteria identified by Zwiren (1989) were applied with caution to the younger participants of these studies. Rowland (1996) suggested that the majority of children in the younger populations might not display a plateau of oxygen uptake as an indication of peak oxygen uptake.

Gentle, but consistent verbal encouragement was given to the participants during the peak oxygen uptake test in order to elicit a maximal effort. A measure of safety for the participants was provided by a member of the research team being positioned behind the exercising participant, at the back of the treadmill at all times during the testing.

3.4.6 Submaximal protocols adopted for the measurement of submaximal O_2 uptakes

3.4.6.1 Discrete Protocols for the Determination of Steady State

Studies One and Two required the participants to run at submaximal running speeds in order to determine a series of steady state oxygen uptakes. Steady state oxygen uptake is defined as the point where there is equilibrium between the oxygen cost of exercise and the oxygen supply to the exercising muscles (Åstrand and Rodahl, 1986). This point is able to be determined metabolically when the oxygen cost of exercise ($\dot{V}O_2$) reaches a plateau after the initial onset of exercise. For the purposes of this thesis, steady state was achieved when the difference in oxygen uptake ($\dot{V}O_2$) in the final two minutes of the exercise bout at submaximal effort was no more than ± 2 ml $kg^{-1}.min^{-1}$. Each participant completed a range of between 4 and 6 discrete submaximal tests. The tests were conducted at submaximal intensities in the range that represented between 30 and 90% of the participant's predetermined peak oxygen uptake. No more than three submaximal exercise bouts were conducted in one day. The length of the submaximal steady state test was six minutes unless otherwise reported in the methodology section of the respective study. The participants also had a recovery interval of at least 20 minutes between determinations.

3.4.7 Continuous incremental protocol for the determination of steady state oxygen uptake on the treadmill and the cycle ergometer

Studies Two and Three also determined steady state submaximal running oxygen uptakes. These studies, however, utilised a continuous non-stop incremental exercise protocol. This continuous incremental protocol dictated that the participants attain steady state at each submaximal workload before progressing to the next workload increment. The definition of steady state described in the preceding section (*Section 3.4.6.1*) was employed.

Each participant completed at least four workload increments in the continuous incremental test. The continuous tests were similar to the discrete exercise tests: they were conducted at

submaximal intensities that represented a range of between 30 and 90% of the participant's pre-determined peak oxygen uptake.

3.4.8 The supramaximal test

Previous studies conducted with a pediatric population have shown that supramaximal tests at intensities that represent 110 – 150% of the subject's peak oxygen uptake show no differences in AOD (Carlson and Naughton, 1993; Buttifant et al., 1996). Therefore the studies in this thesis have used a supramaximal test that represented 120% of peak oxygen uptake. For the purposes of this thesis, the supramaximal test was defined as an exercise bout that was conducted at a constant intensity that was equivalent to 120% of the participant's peak oxygen uptake.

3.4.8.1 Predicting the workload for the supramaximal test

In all studies it was necessary to predict a workload that would elicit a supramaximal effort equivalent to 120% of peak oxygen uptake. In order to do this, individual least square regression equations were constructed from the relationship between submaximal workload and the corresponding oxygen uptake [y = a + b (x)]. The supramaximal workload was then obtained by solving for treadmill speed when the known 120% $\dot{V}O_2$ peak was inserted in the aforementioned calculated linear regression equation (Appendix 3).

3.4.8.1.1 Warm-up Protocol for the supramaximal test

Participants were required to cycle on a Monark™ cycle ergometer with minimal resistance for five minutes prior to the conduct of the supramaximal test. The treadmill was not used because of safety considerations which required at least two members of the research team to be present at the treadmill when a subject was on it. When they had completed the five-minute cycle, they continued to warm up by completing a stretching routine that involved the major muscle groups (quadriceps, hamstrings, calves, gluteals and the lower back), which would be involved in the supramaximal test (Appendix 2.3).

3.4.8.2 Protocol for the supramaximal test conducted on the treadmill

Supramaximal tests were conducted on the treadmill during each of the three studies. At the conclusion of the warm up on the Monark™ ergometer, the participant was asked to stretch the major muscle groups described above (Section 3.4.8.1.1). When satisfied with his warm-up, the participant was requested to straddle the belt on the treadmill. After the all-clear was given, the treadmill was started at the participant's calculated speed. A spotter was positioned directly

behind the participant at the back of the treadmill. The participant was then asked to hold the handrail and raise himself above the treadmill. The spotter supported the participant as he lowered himself onto the treadmill. The spotter continued to support the participant until he had attained the treadmill speed. At that point the test commenced. During the supramaximal test, the participant was vigorously encouraged to run to exhaustion. The test was terminated when the participant reached for the handrail or was given additional support by the spotter.

3.4.8.3 The analysis of expired air from the supramaximal test

Expired air from the supramaximal tests was collected in Douglas bags. Open circuit on-line analysers were not used during this test because expired air was only analysed and calculated at fifteen-second intervals using this equipment. If the test were terminated between the fifteen-second intervals, the data for the intervening time would not be accounted for. Consequently, Douglas bags were used to ensure that all the expired air was collected and analysed. The expired air was collected using a Hans Rudolph one-way valve connected to the Douglas bags via a lightweight tube (with a diameter of 5 cm). Two Douglas bags were attached to a three-way stopcock. Each Douglas bag attachment had its own stopwatch. The stopwatch was activated when the stopcock passed over the start button and opened the Douglas bag to enable expired air collection to take place. When the Douglas bag was full or the test terminated, the stopcock was shifted to close off the Douglas bag. This stopped the stopwatch. In the situation where a Douglas Bag was full, the stopcock device enabled the collection of gas to be transferred into another Douglas Bag.

The Applied Electrochemical analysers described previously were used to determine the concentrations of O_2 and CO_2 of the expired air in the Douglas bag at a rate of 300 $mL.min^{-1}$. The volume of the expired air was determined by a Parkinson Cowan™ ventilometer. The ventilometer was calibrated using a 3-litre calibration syringe (Medical Graphics Corporation). The Douglas bags were evacuated manually. The raw data was then entered into a specifically designed software program based on the Heldane Transformation. These calculations provided measurements of $\dot{V}O_2$ ($L.min^{-1}$ and $mL.kg^{-1}\ min^{-1}$), $\dot{V}_{CO_2}$ ($L.min^{-1}$), $\dot{V}_E$ ($L.min^{-1}$) and the respiratory exchange ratio (RER) for the supramaximal efforts.

3.4.9 Calculating accumulated oxygen deficit

AOD is the difference between the predicted (theoretical) oxygen demand and the actual oxygen uptake for the supramaximal exercise bout. Figure 3.1 illustrates how AOD is calculated.

Figure 3.1 An Example calculation of accumulated oxygen deficit from Supramaximal Test Data.

Participant "John" aged 10 years

Peak oxygen uptake: 1.34 $L.min^{-1}$

Predicted Oxygen Uptake for Supramaximal Test (120%)

1.34 x 1.2 = 1.60 $L.min^{-1}$

Sample of AOD Calculation

Time to exhaustion: (from the supramaximal test)

160.53 seconds (=2.67 min)

Predicted Oxygen Uptake for time:

1.60($L.min^{-1}$) x 2.67 (min)

Σ= 4.27 liters

Actual Oxygen Uptake for Time: (collected during supramaximal test)

1.24 ($L.min^{-1}$) x 2.67 (min)

Σ= 3.31 (liters)

Calculation of Accumulated Oxygen Deficit

Predicted Oxygen Uptake: 4.27 (litres)

Actual Oxygen Uptake 3.31 (litres)

Accumulated Oxygen Deficit = Predicted Oxygen Uptake – Actual Oxygen Uptake

= 4.27 – 3.31 = 0.93 (liters)

3.4.10 Blood collection in Study One

The specific methods employed to collect and analyse blood samples taken from participants in Study One are discussed in the methodology of Chapter 4.

3.4.11 Summary of Protocols

The following table presents a summary of the protocols for each of the studies.

Table 3.2 A summary of the protocols for each of the three studies

Study	Summary of Protocol
One	• peak oxygen uptake test on treadmill • series of discrete submaximal steady state tests • supramaximal test at 120% of peak oxygen uptake
Two A	• peak oxygen uptake test on treadmill • series of discrete submaximal steady state tests • continuous incremental submaximal steady state test • two supramaximal tests at speeds determined by continuous and discrete submaximal tests
Two B	• two continuous incremental submaximal tests on two different days • one series on either the treadmill or the cycle ergometer
Three	• peak oxygen uptake test on treadmill • peak oxygen uptake test on the cycle ergometer • continuous incremental submaximal steady state test on both treadmill and cycle ergometer • supramaximal test at 120% of peak oxygen uptake on both treadmill and cycle ergometer

3.4.12 Overview of the statistical presentation of the data

All data are reported as mean (M) ± standard error of the mean (SE). Differences between groups was analysed using analysis of variance (ANOVA). Newman-Kuels Post Hoc tests were used to locate differences when ANOVA revealed a significant interaction between the groups in Study One. Correlation coefficients were also determined for data in Study One. Student t-tests were used in Study Two B for paired comparisons (Thomas and Nelson, 1996). The SPSS statistical software was used to compute the statistics. The alpha level of 0.05 was accepted as significant. All studies in this thesis were based on an acceptable rejection of the alpha level 0.05; therefore a sample size of 10 would give a detectable power in excess of 80%. A more detailed account of statistical analysis is presented in each experimental chapter.

Chapter 4

STUDY ONE

MEASURING ANAEROBIC PERFORMANCE IN PRE-PUBERTAL, PUBERTAL AND POST-PUBERTAL MALES USING ACCUMULATED OXYGEN DEFICIT

MEASURING ANAEROBIC PERFORMANCE IN PRE-PUBERTAL, PUBERTAL AND POST-PUBERTAL MALES USING ACCUMULATED OXYGEN DEFICIT

Abstract

This study investigated the anaerobic performance of pre-pubertal (age: 11.2 ± 0.4), pubertal (age: 13.7 ± 0.4) and post-pubertal (age: 16.9 ± 0.2) males using accumulated oxygen deficit (AOD). The participants (n = 10 in each group) performed a series of discrete submaximal exercise bouts on the treadmill. Oxygen uptake was measured during the submaximal exercise bouts. Individual linear regression equations were constructed to determine the relationship between submaximal oxygen uptake and exercise intensity. The y-intercept and slope of the individual linear regression lines were used to predict a supramaximal treadmill running speed that equated to a theoretical 120% of the participant's peak oxygen uptake. The participants then ran at the predicted speed on the treadmill. Expired gases were collected in Douglas bags during supramaximal testing. Oxygen uptake was calculated for the supramaximal test and subtracted from the predicted (theoretical) oxygen uptake for the duration of the supramaximal test. This was AOD. A sub-sample of participants (n = 5) in each of the sample groups consented to blood sampling prior to and post supramaximal exercise. Blood samples were analysed for the concentration of blood lactate and pH. One-way analysis of variance (ANOVA) was used to determine whether there were any differences among the three developmental groups. Newman-Kuels Post Hoc analysis was used to locate differences when ANOVA revealed a significant interaction or main effect among the groups. Results from ANOVA tests revealed age-related increments in the absolute AOD ($\sum$L) when the pre-pubertal (1.38 ± 0.21), pubertal (2.88 ± 0.42) and post-pubertal (3.97 ± 0.52) groups were compared ($p < 0.05$). The relative AOD ($\sum mL.kg^{-1}$) of the pre-pubertal (41.37 ± 4.55) group was less than both the pubertal (63.41 ± 7.27) and post-pubertal (54.34 ± 6.78) groups. The post-pubertal group accumulated more blood lactate and produced a greater nadir in pH than both the pubertal and pre-pubertal groups. The findings of this study suggest that the anaerobic performance of different maturational groups can be measured using AOD.

4.1 Introduction

The investigation of the anaerobic performance of children during growth has not received the same amount of research attention as the assessment of aerobic performance (Van Praagh, 1997). Van Praagh (1997) suggested that the comparative lack of research is surprising given the predominance of anaerobic energy used by children on a daily basis. The comparative lack of research may be due to research findings suggesting that the long-term health implications of the cardiorespiratory function of children may have more important health consequences than that of anaerobic function (Van Praagh 1997). Alternatively, it could be argued that anaerobic performance comprises a critical component of competency in sports and physical activity. This is particularly pertinent for children and adolescents in Australia where intermittent based sports are popular.

Anaerobic performances of children during short duration tests ranging between 6 and 30 seconds, have been the most frequently reported (Bar-Or, 1983; Inbar & Bar-Or, 1986; Falgairette *et al.*, 1990; Van Praagh *et al.*, 1990; Medbø, 1991; Mercier *et al.*, 1992; Armstrong *et al.*, 1997; Suei *et al.*, 1998; Doré et al., 2001; Duché et al., 2002; Ratel et al., 2002; Doré et al., 2003; Martin et al., 2003; Martin et al., 2004).

The anaerobic performances of children have shown an age-dependant increase in both peak and mean power in cross-sectional studies (In-Bar and Bar-Or, 1986; Blimkie et al., 1988; Vandewalle et al., 1989; Mercier et al., 1992; Carlson and Naughton 1994). There have been a small number of longitudinal studies examining the anaerobic performance of children using the Wingate Anaerobic Test (Armstrong et al., 2001) and the Force-Velocity Test (Martin et al., 2004). Both studies observed an age dependant increase in peak and mean power.

The examination of anaerobic performance using tests conducted to exhaustion have been relatively few (Carlson & Naughton, 1993; Buttifant *et al.*, 1996; Naughton *et al.*, 1997). Most of the traditional anaerobic tests such as the Wingate Anaerobic Test (WAnT), the force-velocity test and the Sergeant Jump have focused on power output under short-term high-intensity and explosive conditions (Naughton & Carlson, 1998). Some of these short-term tests have been criticised (Saltin, 1990; Withers *et al.*, 1993) for not determining the capacity of the anaerobic system. Accumulated oxygen deficit (AOD) has been proposed as a means of estimating the maximal amount of energy released (the capacity) during a high-intensity effort to exhaustion (Carlson & Naughton, 1998).

AOD remains a controversial measure of anaerobic performance to exhaustion. It is, however, considered to be one of the most acceptable means currently available for measuring anaerobic

performance (Saltin, 1990). The anaerobic performances of adults measured using AOD have been frequently described (Medbø *et al.*, 1988; Medbø & Tabata, 1989; Scott *et al.*, 1991; Bangsbo *et al.*, 1993; Ramsbottom *et al.*, 1994; Pizza *et al.*, 1996; Green & Dawson, 1996a, 1996b). Differences in AOD have been observed between trained and untrained subjects (Scott et al.,1991; Pizza et al., 1996), and adult males and females (Medbø & Burgers, 1990). In addition, AOD demonstrated increases in anaerobic performance with training (Medbø & Burgers, 1990; Tabata *et al.*, 1996; Heugas *et al.*, 1997; Jacobs *et al.*, 1997).

Descriptions of the anaerobic performance of children measured using AOD are limited to comparisons of male and female pre-pubertal participants (Carlson and Naughton, 1993), asthmatic and non-asthmatic pre-pubertal males (Buttifant et al., 1996) and male and female pubertal badminton players (Naughton et al., 1997). Unlike other measures of anaerobic performance in children such as the WAnT and the Force-Velocity Test, where the developmental increase in performance has been well described, AOD, in contrast, has not been investigated in a cross-sectional population of children. Recently, Naughton and Carlson (1998) suggested that one of the future directions for research using AOD with the pediatric population was to examine the developmental aspects of AOD measured from childhood through to adolescence.

4.2 Purpose

The purpose of this study was to use accumulated oxygen deficit to measure anaerobic performance in a cross-section of three groups of male children who represented three developmental stages: pre-pubertal, pubertal and post-pubertal.

4.3 Methodology

This section outlines procedures that are specific to this study only. The methodology of this study will be presented in three sections: general data collection, specific data collection and statistical analysis.

4.3.1 General data collection procedures

Thirty male children representing the developmental stages pre-pubertal (n = 10), pubertal (n = 10) and post-pubertal (n = 10) volunteered to participate in this study. Prior to the commencement of testing, all participants visited the laboratory for a familiarisation session. The nature and purpose of the familiarisation session has been described previously (Chapter 3).

The participants reported to the laboratory on at least four occasions. Laboratory tests required that participant undergo a peak oxygen uptake test, between four and six discrete steady state

oxygen uptake tests and a supramaximal test on a motorised treadmill. These tests were conducted according to the protocol described previously (Chapter 3).

4.3.2 Specific data collection procedures

4.3.2.1 Identifying objective markers of biological maturity

Two means of identifying a participant's level of biological maturity were used during this study. Pubertal staging was obtained via Parent reports of Tanner Scale Stages (Tanner, 1962). Tanner Scale Stages and the concentration of testosterone in saliva were used as objective markers of sexual maturity. Chapter Three describes how parent reporting was conducted.

The participant's level of sexual maturity was seen as important in this cross-sectional study as the actions of the sex hormones, testosterone in particular, are described as critical in growth and development (Winter, 1978). The anabolic sex hormone, testosterone, in particular has been identified as an important contributor to the increased muscle mass observed as a male progresses through puberty (Malina & Bouchard, 1991). Establishing the concentration of testosterone in the participant's body was determined by assessing the concentration of testosterone in saliva. Several authors (Ruutiainen *et al.*, 1987; Vining & McGinley, 1987; Osredkar *et al.*, 1989) have suggested that salivary testosterone is a better indicator of the level of biologically available testosterone in the body than is plasma testosterone. Salivary testosterone is independent of the flow rate of saliva and, when compared with blood sampling (for plasma testosterone), it is a non-invasive and stress-free procedure (Riad-Fahmy *et al.*, 1982). This is especially important when evaluating the testosterone concentrations of children where multiple venepuncture procedures would be extremely stressful.

Testosterone concentration in the body demonstrates a circadian variation. The concentration of testosterone peaks soon after waking and declines during the day to be approximately half the waking concentration in the late evening. Samples collected over several days are also suggested as a way of establishing an accurate mean and representative assessment of basal endocrine activity (Riad-Fahmy et al., 1982).

4.3.2.1.1 Salivary testosterone sampling

In accordance with the recommendations of Raid-Fahmy et al. (1982), participants were asked to provide samples of saliva on three consecutive days. Saliva samples were collected in the morning upon waking, at noon, and in the evening before retiring to bed. Each participant was

given a sealed plastic bag containing nine labeled 5-ml plastic sample tubes. Instructions about how to provide the salivary sample were written on the bag. These instructions were:

1. Collect the sample in the morning, at lunchtime and in the evening
2. Collect samples for three consecutive days
3. Wash the mouth out with clean water before collecting the sample
4. Label the sample with the date and time
5. Store the sample in the freezer until all samples have been collected.

After the participant had collected all of the samples, they were asked to return them to the laboratory where they were stored at –20°C for subsequent analysis. This sampling regime was cognisant of the diurnal nature of testosterone and subsequently established a base line of endocrine activity.

4.3.2.1.2 Salivary testosterone analysis

Salivary testosterone samples were analysed by radioimmunoassay using a DPC Coat – A-Count kit (TTKTT1). Prior to analysis, the saliva samples were thawed and centrifuged for 10 minutes. The resultant salivary supernatant was then transferred to a sterile container using a transfer pipette. The DPC Coat-A-Count kit was modified according to the manufacturer's instructions for use with salivary samples as it had been originally designed for use with serum or urine samples. The modification required the standards provided in the kit to be diluted 1:20 with milli Q water. This was done by adding 50μL of each standard to 950μL of milli Q water. The quality control was also diluted in the same manner. The standards and the quality controls were then mixed by gentle vortexing. The next step was to pipette 200μL of the diluted standards and quality controls into the previously labelled tubes. The same amount of undiluted salivary samples were then pipetted into tubes labelled with the participant's identification number and the sample number. Testosterone [125] was subsequently added to all of the tubes in 1.0ml aliquots. The tubes were then vortexed and covered with clear plastic wrap. Incubation took place overnight (16-24 hours) at room temperature. The samples were decanted, following incubation. This procedure involved tipping the contents of the tubes into a sink specifically designated as suitable for disposing of radioactive fluids. Tap water was running into the sink throughout the tipping procedure in order to clear the radioactive products as thoroughly as possible. After the liquid was poured from the tubes, they were vigorously shaken. The tubes were then turned upside down on an absorbent pad for 2 to 3 minutes to drain any excess moisture. The removal of all visible moisture from the

tubes greatly enhanced the precision of the assay, so the tubes were also vigorously shaken on the absorbent pad to dislodge any remaining droplets. Following this procedure, the tubes were then loaded into the gamma counter sampling racks in the appropriate order. The samples were counted for one minute and at the conclusion of the counting process the computer attached to the gamma counter printed the results. The salivary samples were recorded in pico mol/litre (pmol.l^{-1}).

4.3.3 Blood collection and metabolite analysis

Pre-pubertal children respond differently to high intensity exercise when compared with their more mature counterparts (Eriksson & Saltin, 1974; Inbar & Bar-Or, 1986; Van Praagh, 2000). It is generally reported that pre-pubertal children have lower peak lactic acid concentration after maximal exercise compared with pubertal and post-pubertal children (Eriksson *et al.*, 1973; Eriksson & Saltin, 1974; Eriksson, 1979; Van Praagh *et al.*, 1989; Falgairette *et al.*, 1990; Pianosi *et al.*, 1995; Pfitzinger & Freedson, 1997). Pre-pubertal children do not experience the same degree of acidosis (Matejkova *et al.*, 1980) after maximal exercise when compared with their more mature counterparts.

The cross-sectional design of the study provided an opportunity to examine the potential differences in the metabolic responses of pre-pubertal, pubertal and post-pubertal groups to the supramaximal exercise bout. A sub-sample of participants (n = 5) from each of the groups consented to providing blood at rest and at minutes 1, 3, 5, 7, 10, 15, and 20 minutes post exercise. The blood samples were analysed for blood lactate concentration [La$^-$], plasma pH and haemoglobin concentration [Hb].

4.3.3.1 Blood sampling

A trained and experienced member of the research team inserted an indwelling catheter into a forearm vein while the participant was resting supine. The catheter was then fixed with a three-way stopcock to enable serial blood sampling. Each participant's hand was also warmed in a water bath in order to stimulate arterial blood. Two blood samples were drawn. Initially a 2 ml sample was withdrawn and discarded. Then 2 ml of blood was extracted for blood gas analysis. A further 5 ml of blood was sampled for subsequent analysis of blood [La$^-$]. After blood sampling was completed, a further 1 ml of heparinised saline was injected into the catheter to maintain potency during exercise testing.

Immediately after the conclusion of the supramaximal test, the participant was instructed to move from the treadmill and lie down on a portable bed. The participant's hand was then placed in a

warm water bath. At 1-minute post exercise, 7 ml of blood (2 ml + 5 ml) was drawn after the catheter was initially cleared in the manner previously described. Additional blood sampling occurred at minutes 3, 5, 7, 10, 15, and 20-post exercise.

4.3.3.2 Preparation of blood samples

After the 5 ml blood sample was collected, it was slowly injected into a chilled lithium heparin tube and gently rolled to avoid coagulation. It was then stored in ice for subsequent treatment.

4.3.3.2.1 Blood Lactate

A 500µl aliquot of the stored anticoagulated blood was pipetted into an eppendorf tube containing 1ml of ice-cold 3M PCA. The sample was then vortexed and centrifuged for 2 minutes at 2000 rpm. The resultant supernatant was poured into a labelled eppendorf tube and stored at –80°C until analysed for blood lactate. Blood lactate concentrations were determined spectrophotometrically (Shimadzu UV-120) using an enzymatic technique (Lowry & Passonneau, 1972).

4.3.3.2.2 Blood Gas, Hb, and pH Analysis

A 2 ml sample was drawn into a syringe containing heparin (5000 IU/ml). From this sample, pH and Hb were determined using an automated blood gas analyser (Radiometer, Copenhagen, ABL-30 acid/base analyser). All samples were analysed in duplicate.

4.3.4 Statistical analysis

The descriptive data was expressed as means ± standard errors (Mean ± S.E.M.). Linear regression equations were constructed to describe the relationship between submaximal oxygen uptake and exercise intensity. A series of one-way analyses of variance (ANOVA) tests were conducted to determine the differences between the three developmental groups. Newman-Kuels Post Hoc analysis were used to locate differences when an ANOVA revealed a difference or main effect between the groups (SPSS Version 5). Scheffè's method was used to examine how quickly the blood borne metabolites returned to resting levels after exercise. The alpha level of 0.05 was accepted as the level of probability with which to reject the null hypothesis.

4.4 Results

The focus of this study was to examine the anaerobic performance characteristics in a cross-section of pre-pubertal, pubertal and post-pubertal males using accumulated oxygen deficit (AOD). The results of this study will be presented in three major sections: participant profiles, accumulated oxygen deficit measures, and blood parameter responses to anaerobic performance.

4.4.1 Participant profiles

4.4.1.1 Descriptive characteristics

The descriptive characteristics of the sample groups are presented in Table 4.1. The post-pubertal group was significantly ($p < 0.05$) heavier and taller than either the pre-pubertal and pubertal groups (Appendix 4.1). In percentage terms, the post-pubertal group was 52% heavier and 20% taller than the pre-pubertal group. When compared with the pubertal group, the post-pubertal group was 33% heavier and 7% taller. The pubertal group was 27% heavier and 13% taller when compared with the pre-pubertal group.

The pre-pubertal and pubertal groups were of average mass and height when compared with the percentile curves for caucasian males of the same age. The post-pubertal group was slightly heavier (75^{th} percentile) and marginally taller (60^{th} percentile) when compared with the percentile curves for caucasian males of the same age (National Health and Nutrition Examination Survey (NHANES), 1992). The differences in mass (kg) and height (cm) of the participants' of the study group would be expected given the rate of growth that occurs in children between pre and post puberty.

4.4.2 Maximal effort profiles

The maximal effort profiles of the participants who participated in this study are presented in Table 4.2. There were differences ($p < 0.05$) in the absolute ($L.min^{-1}$) measures of peak oxygen uptake when the post-pubertal group was compared with both the pre-pubertal and pubertal groups (Appendix 4.1). The post-pubertal group produced a 58% greater peak oxygen uptake ($L.min^{-1}$) when compared with the pre-pubertal group. A similar trend was evident when the post-pubertal group was compared with the pubertal group. The post-pubertal group exhibited a 40% greater peak oxygen uptake ($L.min^{-1}$) than the pubertal group. The pubertal group produced a 29% greater peak oxygen uptake ($L.min^{-1}$) when compared with the pre-pubertal group. The increase in peak oxygen uptake ($L.min^{-1}$) observed with varying pubertal status in this study

closely parallels the growth and maturation of the pulmonary, cardiovascular and peripheral determinants of peak oxygen uptake ($L.min^{-1}$) (Rowland, 1996).

Relative peak oxygen uptake ($mL.kg^{-1}.min^{-1}$) also showed differences ($p< 0.05$) when the three groups were compared (Appendix 4.1). The percentage differences in relative ($mL.kg^{-1}.min^{-1}$) peak oxygen uptake of the groups were comparatively smaller than the differences observed in absolute ($L.min^{-1}$) peak oxygen uptake. The post-pubertal group exhibited a 14% greater peak oxygen uptake when compared with the pre-pubertal group. When compared with the pubertal group the post-pubertal group had a 10% higher relative ($mL.kg^{-1}.min^{-1}$) peak oxygen uptake. A much smaller percentage difference (4%) was evident when the pre-pubertal group was compared with the pubertal group.

Table 4.1 Descriptive characteristics of the groups.

	Group		
Variable	**Pre-pubertal (n = 10)**	**Pubertal (n = 10)**	**Post-pubertal (n = 10)**
Age	11.2 [a,b]	13.7 [c]	16.9
(yr)	± 0.4	± 0.4	± 0.2
Mass	35.35 [a,b]	48.58 [c]	72.78
(kg)	± 2.34	± 2.38	± 1.89
Height	143 [a,b]	164 [c]	177
(cm)	± 2.77	± 2.98	± 2.02

Mean ± S.E.M.

'a' denotes pre-pubertal different from pubertal ($p< 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p< 0.05$)

'c' denotes pubertal different from post-pubertal ($p< 0.05$)

All of the participants achieved at least one of the criteria for maximal performance suggested by (Zwiren, 1989). These criteria include: peak heart rates that are 95% of the age-predicted maximum; a levelling-off of oxygen uptake with increasing workload; or a respiratory exchange ratio (RER) in excess of 1.0 at peak oxygen uptake. There was a difference ($p< 0.05$) in the

maximal heart rate achieved at peak oxygen uptake when the pre-pubertal and pubertal groups were compared with the post-pubertal groups (Appendix 4.1).

4.4.3 Measures of biological maturity

The selected markers of biological maturity, namely the Tanner Scale (Tanner, 1962) scores for pubic hair and salivary testosterone concentration (pmol), together qualified the differences in the stages of biological maturity between the groups. Mean concentrations of salivary testosterone (pmol) are presented in Figure 4.1. The mean scores for the Tanner Scale (Tanner, 1969) were 1, 3 and 5 for the pre-pubertal, pubertal and post-pubertal groups, respectively. These scores were different ($p < 0.05$) from each other (Appendix 4.2). The mean concentrations of testosterone in saliva showed a similar trend to those of the Tanner Scale scores with the post-pubertal group exhibiting a higher salivary testosterone concentration (pmol) ($p < 0.05$) when compared with both the pubertal and pre-pubertal groups (Appendix 4.2). The pre-pubertal group produced only 0.09% of that produced by the post-pubertal group and 18% of that of the pubertal group. The pubertal group produced 51% of that produced by the post-pubertal group.

Table 4.2 Maximal effort profiles

	Group		
Variable	**Pre-pubertal (n = 10)**	**Pubertal (n = 10)**	**Post-pubertal (n = 10)**
Peak $\dot{V}O_2$ ($L.min^{-1}$)	2.12 [a,b] ±0.13	2.99 [c] ± 0.18	4.99 ± 0.19
Peak $\dot{V}O_2$ ($mL.kg^{-1}.min^{-1}$)	59.41 [b] ± 2.27	61.99 [c] ± 1.97	68.86 ± 2.76
Peak Heart Rate ($b.min^{-1}$)	202 [b] ± 1	201 ± 1	194 ± 2

Mean ± SEM.

'a' denotes pre-pubertal different from pubertal ($p < 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

'c' denotes pubertal different from post-pubertal ($p < 0.05$)

4.4.4 Accumulated oxygen deficit measures

The following section presents results of mean submaximal exercise intensities, mean linear regression equations, AOD measures and relative contributions of the aerobic and anaerobic systems to the supramaximal test for each of the pre-pubertal, pubertal and post-pubertal groups.

4.4.4.1 Submaximal exercise intensity comparisons

Submaximal steady state exercise was conducted on a motorised treadmill as previously described. Each participant completed at least four submaximal steady exercise bouts at speeds ranging from 6 – 12 $km.hr^{-1}$. Table 4.3 presents the mean values for the lowest exercise intensity and the highest exercise intensity expressed as a percentage of the peak oxygen uptake ($mL.kg^{-1}.min^{-1}$) for each of the groups. The range of the lowest and highest submaximal exercise intensities, for the total sample group, were 40.5 – 77.5 and 60.7 – 91.8 for the lowest and highest exercise intensities respectively.

Table 4.3 Mean highest and lowest submaximal steady state intensities (% $\dot{V}O_2$ $ml/kg^{-1}.min^{-1}$)

	Group		
Variable	**Pre-pubertal (n = 10)**	**Pubertal (n = 10)**	**Post-pubertal (n = 10)**
Mean lowest % $\dot{V}O_2$	58.7 ± 3.7	59.6 ± 3.4	56.0 ±3.1
(range)	(40.8 – 77.5)	(43.6 – 76.7)	(46.6 – 73.5)
Mean highest % $\dot{V}O_2$	82.5 ± 1.9	77.5 ± 2.4	74.1 ± 2.9
(range)	(73.1 – 90.8)	(66.5 – 91.9)	(60.7 – 86.1)

Mean ± S.E.M.

There were no differences ($p > 0.05$) in the mean lowest or highest submaximal exercise intensities when the groups were compared (Appendix 4.3). Medbø and Tabata (1989) proposed that steady state submaximal exercise be conducted at intensities that represented 30 - 90% of the participant's peak oxygen uptake. The mean lowest and highest submaximal exercise intensities fell within this range (Medbø and Tabata, 1989).

4.4.4.2 Mean linear regression equations

Subsequent to the conduct of the submaximal steady state tests, individual regression analyses were conducted. The regression analyses were of linear form [y = a + b(x)] and described the relationship between submaximal oxygen uptake and exercise intensity. In order to predict the supramaximal workload, the linear regression equation was rearranged so that [y = a + b(x)] read (x) = (y - a)/b where y represented oxygen uptake and (x) represented the exercise intensity (see Appendix 3 for an example calculation). The resultant Y intercept (a) and slope of the regression line (b) are used to predict the supramaximal workload that represents 120% of the participants' maximal oxygen uptake. The mean linear regression equations for each of the groups are presented in Table 4.4.

There were no statistically significant differences ($p > 0.05$) in any of the variables that describe the relationship between submaximal oxygen uptake and exercise intensity (Appendix 4.4).

A notable difference is the Pearson correlation coefficient (r) of each of the groups. The Pearson correlation coefficient for the post-pubertal group was 6% lower than that of the pre-pubertal group and 4% lower than that of the pubertal group when the values were compared. The accountable variance represents the variance in the confidence in the linearity of the regression line. These regression lines express the relationship between submaximal oxygen uptake and exercise intensity; therefore the accountable variance represents the variance in submaximal oxygen uptake that can be accounted for by the submaximal exercise intensity. In the pre-pubertal group, 96% of the variance in submaximal oxygen uptake is accounted for by the exercise intensity. The accountable variance of the pubertal group is 92% and in the post-pubertal group the accountable variance is 85%. When compared with the pre-pubertal group, the confidence in the linearity of the regression line is less in the post-pubertal group.

The linear regression equations were then used to predict the exercise intensity that represented 120% of the participants' peak oxygen uptake (supramaximal exercise). As treadmill running was the mode of exercise for this study, treadmill speeds were predicted from the individual linear regression equations. For an example of this calculation, see Appendix 3. The mean treadmill speed required to elicit 120% of peak oxygen uptake for the pre-pubertal group was 12.58 ± 0.80 $km.hr^{-1}$, the pubertal group 14.80 ± 0.73 $km.hr^{-1}$ and the post-pubertal group 17.23 ± 0.91 $km.hr^{-1}$. The post-pubertal speed was statistically greater ($p = 0.02$) than the treadmill speeds predicted for the pre-pubertal and pubertal groups (Appendix 4.5).

Table 4.4 Mean linear regression equations

	Group		
Variable	**Pre-pubertal (n = 10)**	**Pubertal (n = 10)**	**Post-pubertal (n = 10)**
Y intercept	0.86 ± 0.41	0.63 ± 0.37	-1.77 ± 1.47
Slope	0.15 ± 0.0	0.17 ± 0.0	0.18 ± 0.1
Pearson Correlation (r)	0.98	0.96	0.92
Accountable Variance (R^2) (r^2) x 100	96%	92%	85%

Mean ± S.E.M.

4.4.5 Measures of accumulated oxygen deficit

Anaerobic performance characteristics measured by AOD are presented in Table 4.5 and Figures 4.2 and 4.3. There were differences ($p < 0.05$) between each of the three groups when AOD was expressed in litres (ΣL) (Appendix 4.6). The percentage differences in the absolute (ΣL) AOD of the pre-pubertal group when compared with the pubertal and post-pubertal groups were 52% and 65%, respectively. A much smaller percent difference (27%) was observed between the pubertal group and the post-pubertal group. ANCOVA was conducted to remove the effect of the differences in height and mass of the groups. ANCOVA revealed that there were still differences when the effect of height and mass were statistically removed (Appendix 4.6). When AOD values are presented in relative terms ($\Sigma mL.kg^{-1}$) there was a difference ($p < 0.05$) in the pre-pubertal group (41.37 ± 3.55) when compared with the pubertal group (63.41 ± 7.27) and the post-pubertal group (54.43 ± 6.78). There were no differences in AOD ($\Sigma mL.kg^{-1}$) between the pubertal and post-pubertal groups (Appendix 4.6). The finding of no difference in the AOD ($\Sigma ml.kg^{-1}$) when the pubertal and post pubertal groups were compared may be due to the low effect size (Appendix 4.7) when the groups were compared.

The percentage differences in the relative ($\Sigma mL.kg^{-1}$) AOD of the pre-pubertal group were smaller than those observed for the absolute AOD (ΣL) when compared with the pubertal group

(30%) and the post-pubertal group (24%). The pubertal group exhibited an 8% larger relative ($\Sigma mL.kg^{-1}$) AOD than the post-pubertal group.

The analysis of other variables of the supramaximal performance; test time, supramaximal heart rate, and RER revealed no statistically differences ($p > 0.05$) between any of the groups (Appendix 4.6).

Table 4.5 Anaerobic performance characteristics

	Group		
Variable	**Pre-pubertal (n = 10)**	**Pubertal (n = 10)**	**Post-pubertal (n = 10)**
AOD (ΣL)	1.38 ± 0.21 [a b]	2.88 ± 0.42 [c]	3.97 ± 0.52
AOD ($\Sigma mL.kg^{-1}$)	41.37 ± 4.55 [a b]	63.41 ± 7.27	54.43 ± 6.78
Heart Rate ($b.min^{-1}$)	193 ± 2	194 ± 2	186 ± 3
Test Time (sec)	148.54 ± 18.67	175.17 ± 35.39	120.34 ± 23.01
RER	1.08 ± 0.02	1.12 ± 0.02	1.14 ± 0.02

Mean ± S.E.M.

'a' denotes pre-pubertal different from pubertal ($p < 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

'c' denotes pubertal different from post-pubertal ($p < 0.05$)

4.4.6 Relative aerobic and anaerobic contributions to the supramaximal test

The relative aerobic and anaerobic contributions (%) to the supramaximal exercise bout were estimated using accumulated oxygen demand. This was achieved by dividing the actual oxygen consumed during the supramaximal exercise bout by the predicted accumulated oxygen demand. This gave an estimation of the relative aerobic contribution to the exercise bout.

The post-pubertal group was estimated to have an anaerobic contribution to the supramaximal exercise bout of 32%. The pubertal and pre-pubertal groups were estimated to have a 28% and 22% anaerobic contribution to the supramaximal exercise bout, respectively. These estimations represent a 10% and 4% greater anaerobic contribution to the supramaximal exercise bout from the post-pubertal group when compared with the pre-pubertal and pubertal groups, respectively. Accumulated oxygen demands for the supramaximal tests are presented in Figure 4.4.

4.4.7 Blood parameter responses to supramaximal exercise

A sub-group of pre-pubertal (n = 5), pubertal (n = 5) and post-pubertal (n = 5) participants consented to providing blood samples prior to and post supramaximal exercise. The number of pre-pubertal participants who consented to the cannulation procedure and the subsequent blood sampling limited the sample size to five in each group. The following section presents the results of pH, blood lactate measures of the participants to supramaximal exercise.

4.4.7.1 pH responses to supramaximal exercise

Figure 4.5 illustrates the pH response following the supramaximal test. The post-pubertal group experienced a larger change in pH concentration from rest to 1 minute post exercise of 0.21 units. The pubertal group experienced a change of 0.14 units. The pre-pubertal group experienced the smallest change of 0.12 units.

There were no differences ($p > 0.05$) in the pH values between the groups prior to exercise (Appendix 4.8). The range of pre-exercise plasma pH values 7.32 to 7.44, 7.32 to 7.42 and 7.36 to 7.42 for the pre-pubertal, pubertal and post-pubertal groups, respectively. The pre-pubertal group experienced its mean nadir for plasma pH (7.22 ± 0.01) at 1-minute post supramaximal exercise. At this point there were no differences in the pH of all of the groups ($p > 0.05$) (Appendix 4.8). Three minutes after the supramaximal test, the pubertal and post-pubertal groups experienced their mean plasma pH nadir. Mean nadir in plasma pH of 7.22 ($\pm$ 0.03) and 7.17 ($\pm$ 0.03) were observed for the pubertal and post-pubertal group, respectively. The post-pubertal group

experienced greater acidosis when compared with the pre-pubertal group at minutes 3, 5, 7, 10 and 20-post exercise ($p < 0.05$) (Appendix 4.8).

An analysis of the between subject effects for each of the groups (Appendix 4.8.1) demonstrated a different time course in the recovery of plasma pH after the supramaximal test. The pre-pubertal group exhibited a significant increase in plasma pH from minutes 5 to 7 and then again at minutes 10 to 15. Both the pubertal and post-pubertal groups failed to demonstrate an increase in plasma pH until minutes 7 to 10. The post-pubertal group continued to show increases in pH from 7 to 20 minutes following the supramaximal test ($p < 0.05$). The trend in the pH response of the pubertal group differed from that of the post-pubertal group (Figure 4.5).

4.4.7.2 Blood lactate responses to supramaximal exercise

The concentrations of blood lactate for each of the groups are presented in Figure 4.5. The mean concentrations of blood lactate prior to exercise were 1.00 (± 0.00), 0.66 (± 0.18) and 1.08 (± 0.31) $mM.l^{-1}$ for the pre-pubertal, pubertal and post-pubertal groups, respectively.

The post-pubertal group experienced the greatest post- exercise change in blood lactate (Δ 7.44 units) at 1 minute exercise. The pubertal and pre-pubertal groups experienced a smaller change of 4.39 and 4.56 units, respectively. In contrast to the other groups, the pubertal group experienced another increase in blood lactate concentration of 0.4 units from minutes 1 to minute 3 post-exercise.

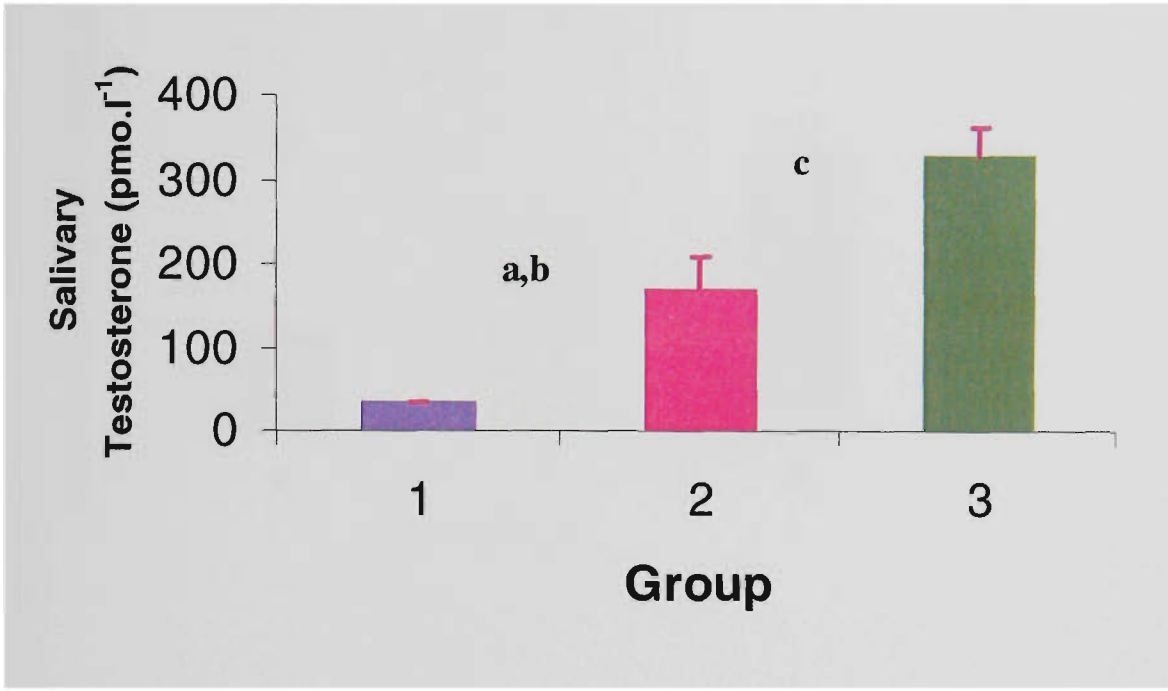
The increase in the concentration of lactic acid in the blood followed the same time course as plasma pH. There was, however, a difference ($p < 0.05$) in the blood lactate concentration of the pre-pubertal group when compared with the post-pubertal group from 1-minute post supramaximal exercise. Blood lactate concentration at minute 1 also showed a difference between the pubertal (4.96 ± 0.53 $mMl.^{-1}$) and post-pubertal (8.48 ± 1.27 $m.Ml.^{-1}$) groups ($p = 0.34$). The pubertal group did not reach a peak blood lactate concentration until minute 3 (Appendix 4.9).

An analysis of the between participant differences in the blood lactate concentration over time was conducted for each of the groups (Appendix 4.9.1; Figure 4.5). The pre-pubertal group demonstrated a decrease ($p < 0.05$) in blood lactate concentration between minutes 3 and 5 and again between minutes 5 to 7. A further decrease occurred between minutes 15 and 20. In contrast, the concentration of blood lactate of the pubertal and post-pubertal groups did not decrease until between minutes 5 and 7. A decrease in the blood lactate concentration in the pubertal group was again evident between minutes 7 and 10 and minutes 15 to 20. In contrast, the

concentration of blood lactate in the post-pubertal group continued to decrease from minute 7 until blood sampling was concluded at 20 minutes post supramaximal exercise.

Hemoglobin levels were measured prior to the supramaximal exercise bout. At rest, there were differences in haemoglobin concentration (g/dl) when the pre-pubertal group (12.56 ± 0.27) was compared with the pubertal (14.05 ± 0.69) and the post-pubertal group (14.78 ± 0.15) (Appendix 4.10).

Figure 4.1 Mean salivary testosterone concentration ($pmol.l^{-1}$)



Mean ± S.E.M.

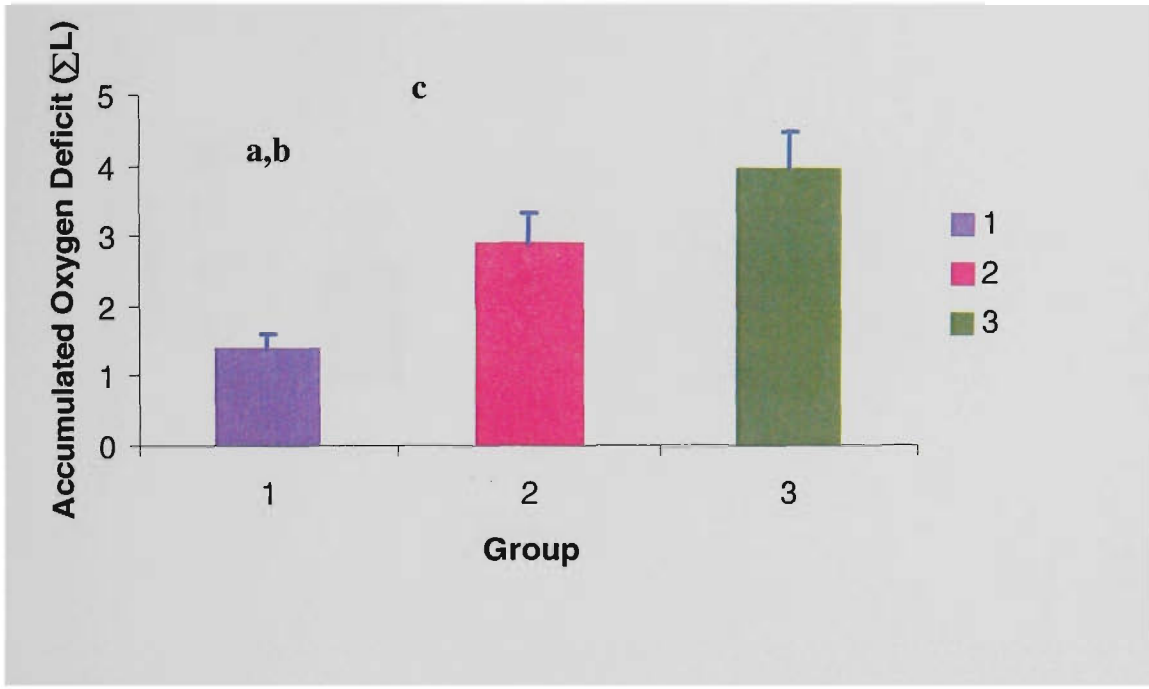
1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal.

'a' denotes pre-pubertal different from pubertal ($p < 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

'c' denotes pubertal different from post-pubertal ($p < 0.05$)

Figure 4.2 Mean accumulated oxygen deficit (∑L)



Mean ± S.E.M.

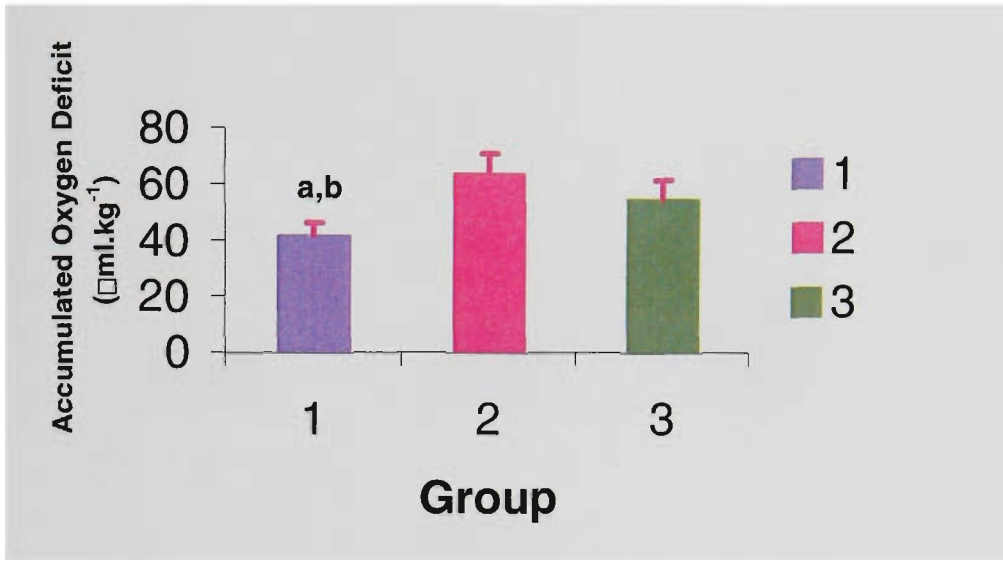
1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal

'a' denotes pre-pubertal different from pubertal ($p < 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

'c' denotes pubertal different from post-pubertal ($p < 0.05$)

Figure 4.3 Mean accumulated oxygen deficit ($\Sigma mL.kg^{-1}$)



Mean ± S.E.M.

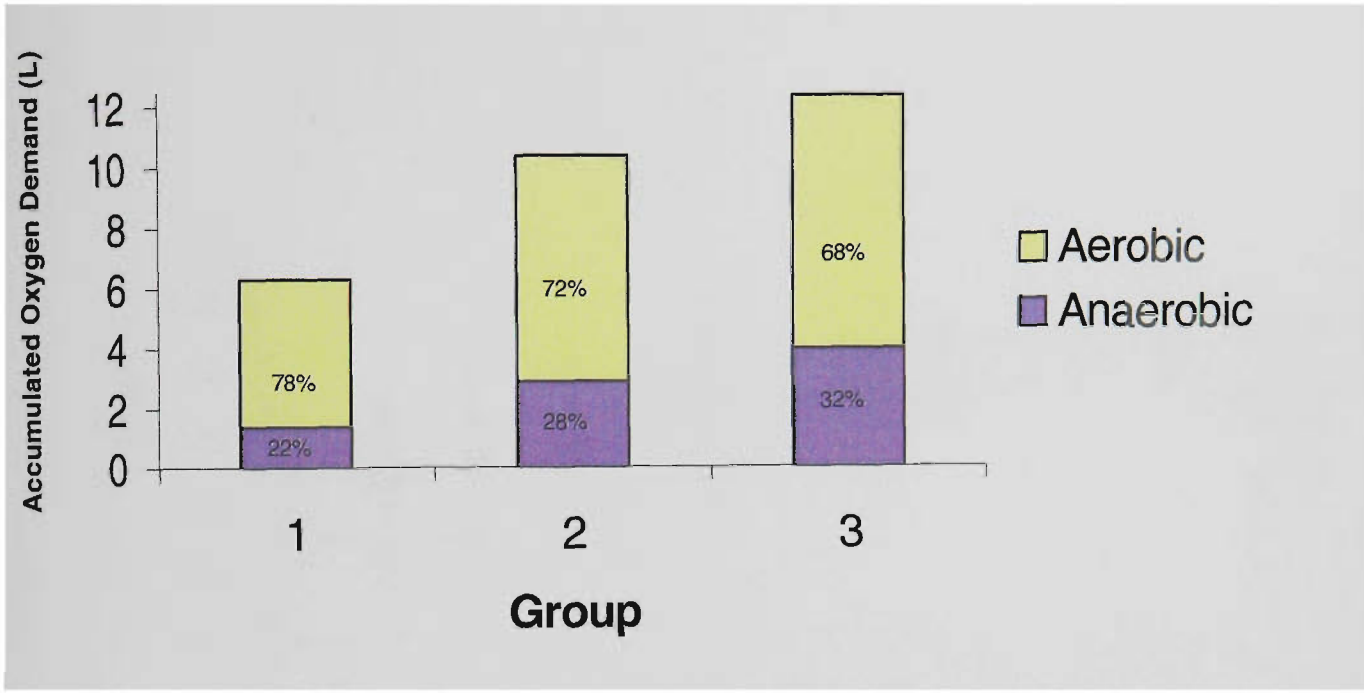
1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal.

'a' denotes pre-pubertal different from pubertal ($p < 0.05$)

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

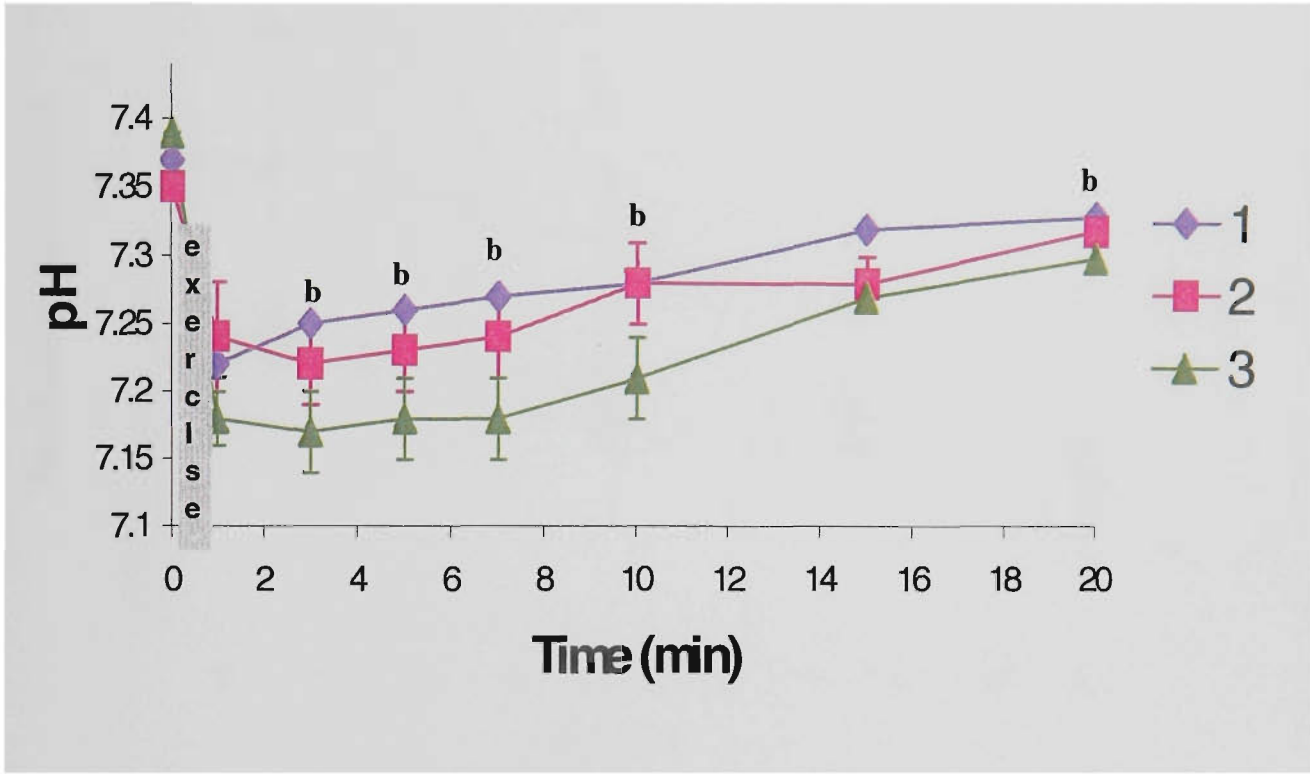
'c' denotes pubertal different from post-pubertal ($p < 0.05$)

Figure 4.4 Relative aerobic and anaerobic contributions (%) to the supramaximal test



1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal.

Figure 4.5 pH responses to supramaximal exercise

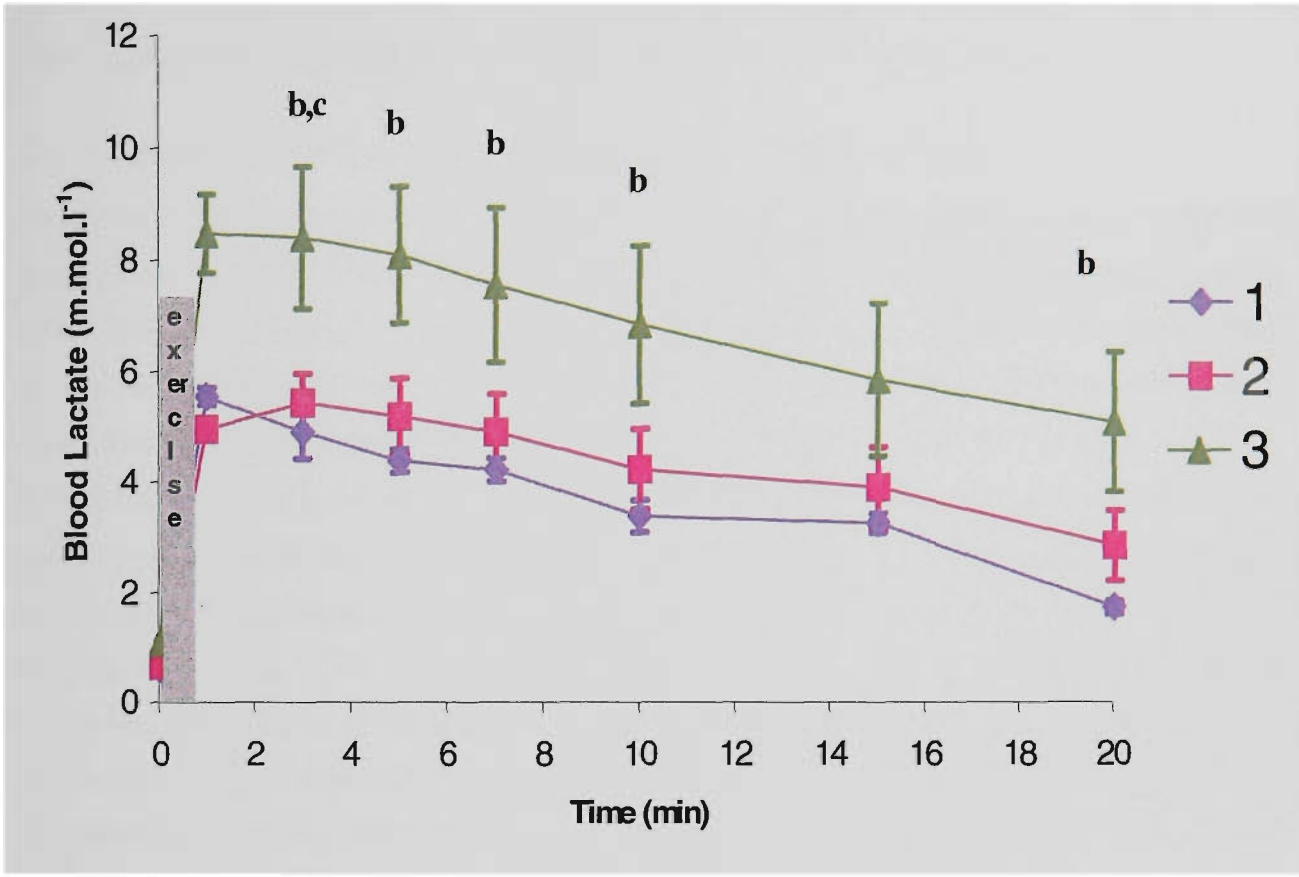


Mean ± S.E.M.

1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal.

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

Figure 4.6 Blood lactate responses to supramaximal exercise



Mean ± S.E.M.

1 - pre-pubertal; 2 - pubertal; 3 - post-pubertal.

'b' denotes pre-pubertal different from post-pubertal ($p < 0.05$)

'c' denotes pubertal different from post-pubertal ($p < 0.05$)

4.5 Discussion

This study investigated the anaerobic performance of a cross-section of males representing pre-pubertal, pubertal and post-pubertal stages of development. The measurement of AOD in absolute terms (ΣL) revealed differences ($p < 0.05$) between all of the maturational groups. AOD measured in relative (ΣmL.kg^{-1}) terms revealed differences ($p < 0.05$) between the pre-adolescent group when compared with the pubertal and post-pubertal groups only. There were no differences between the pubertal and post-pubertal groups when AOD (ΣmL.kg^{-1}) was compared.

4.5.1 Accumulated Oxygen deficit: Comparison among the groups

There were differences ($p < 0.05$) in AOD when the pre-pubertal, pubertal and post-pubertal groups were compared. This difference may be related to the physical growth of the participants. When indicators of physical growth such as mass and height are compared, the post-pubertal group were significantly heavier and taller than both the pre-pubertal and pre-pubertal groups. There was also a strong correlation between mass ($r = 0.686$, $p = 0.000$) and height ($r = 0.695$, $p = 0.000$) and AOD (ΣL). In addition to physical growth indicators that were not measured in this study, factors such as active muscle mass may have contributed to the differences in AOD (ΣL). A 19.4% increase in muscle mass in boys between the ages of 10 and 16 years has been observed (Rassmussen *et al.*, 1990). The mass of muscle contributes to body mass and the pre-pubertal group was not as heavy as the pubertal or post-pubertal groups. One could conclude that the size of the active muscle mass could also be smaller in the pre-pubertal group when compared with the pubertal and post-pubertal groups. Medbø (1991) commented on the size of the active muscle mass in relation to AOD (ΣL) and suggested that the subjects with the largest AOD (ΣL) may have the largest active muscle mass. The relationship between the smaller AOD (ΣL) in the pre-pubertal group when compared with the pubertal and post-pubertal groups and active muscle mass, is supported by findings of a strong correlation ($r = 0.94$, $p < 0.001$) between lean body mass and anaerobic power in 11 to 19 year old boys (Mercier *et al.*, 1992). Although AOD and measures of anaerobic power using the force-velocity test are different measures of anaerobic performance, comparisons between these measures of anaerobic performance are useful.

Qualitative differences within the muscle could also provide an explanation for the smaller AOD (ΣL) of the pre-pubertal group when compared with the pubertal group, and when the pubertal group were compared with the post-pubertal group. Studies of the anaerobic metabolism of children have reported that the anaerobic metabolism of children is lower than that of adults (Eriksson *et al.*, 1973; Paterson *et al.*, 1986; Zanconato *et al.*, 1993). Zanconato et al. (1993)

using Phosphorous 31 nuclear magnetic resonance spectroscopy (^{31}P NMRS) reported a different response in the inorganic phosphate (Pi)/phosphocreatine (PCr) and pH during high-intensity exercise. The different response in the Pi/PCr ratio suggests that when the exercise intensity reached a certain level, pre-pubertal children were no longer able to provide energy for exercise through anaerobic metabolism. This observation is supported by findings of a smaller pH drop in the pre-pubertal children when compared with the adults in the study (Zanconato, et al., 1993). The minimal decrease in pH provides additional support to the theory that pre-pubertal children are less able to provide energy through anaerobic metabolism. The authors concluded that there might be growth-related differences in anaerobic energy metabolism that limit pre-pubertal children's ability to exercise at high-intensities (Zanconato et al., 1993). Kuno et al., (1995) also support these findings in a ^{31}P NMRS study investigating the muscle metabolism of 12 - 15-year-old males. The smaller anaerobic capabilities of the pre-pubertal (Zanconato et al., 1993) and the pubertal (Kuno *et al.*, 1995) subjects when compared with adults support the findings of the present study with respect to the smaller anaerobic performances of the pre-pubertal and pubertal participants when compared with the post-pubertal participants.

4.5.2 Accumulated oxygen deficit ($\Sigma mL.kg^{-1}$): Comparisons among the groups

There was a difference in AOD between the pre-pubertal group when compared with the pubertal and post-pubertal groups only. There was no difference in AOD when the pubertal and post-pubertal groups were compared. These findings may be due to the different training status of the two groups and the effect of growth. AOD has been used to show improvements in anaerobic performance with training (Medbø and Burgers, 1990; Jacobs et al., 1996; Tabata et al., 1996; Heugas et al., 1997). In support of the observations of an increased AOD with training in adults, Naughton and Carlson (1998) suggested that anaerobic performances measured by AOD might be reflective of exercise training during the pubertal years. AOD may, therefore, detect differences in the training status of the pubertal and post-pubertal groups. The effect of training status may be further impacted upon by the differences in growth in the pubertal group in particular. Of each of the three groups studied, the pubertal group represented the greatest range in growth. The biological range of the salivary testosterone concentrations and the range of Tanner Scale scores in this study, support this notion. Participants in the pubertal group ranged from Tanner Stage 2 through to Tanner Stage 4. Their salivary testosterone concentrations ranged from 59 – 375 $pmol.l^{-1}$. The salivary testosterone concentrations of the pubertal group encompassed a much greater biological range when compared with the post-pubertal group (259 – 552 $pmol.l^{-1}$) who exhibited adult concentrations. The pubertal group was a less homogeneous group in terms of

their growth when compared with the post-pubertal group, which exhibited more homogeneous growth characteristics. The possible impact of the differences in the growth of this group could have been compounded by their training status. Both of these factors could impact on AOD ($\Sigma mL.kg^{-1}$). Inbar and Bar-Or (1983) and Mercier et al., (1992) support this notion, stating that it is sometimes difficult to differentiate between the effects of growth and training in pediatric research. This notion is further confirmed by the findings of Naughton et al. (1997). Naughton and her colleagues investigated the anaerobic performance of a group of anaerobically trained pubertal males and females using AOD. AOD values of 71.5 $mL.kg^{-1}$ in pubertal males at 120% of peak oxygen uptake were reported. These AOD values are greater than some values reported for untrained (Medbø et al., 1988; Medbø and Burgers, 1990; Scott et al., 1991), endurance-trained (Hermansen and Medbø, 1984; Medbø and Burgers, 1990; Scott et al., 1991) and sprint-trained adult males (Hermansen & Medbø, 1984). The effect of training, in the present study, may be greater than the effect of growth when AOD is used to measure anaerobic performance.

4.5.3 Accumulated oxygen deficit: Comparisons with previous studies in the pediatric population.

AOD has previously been measured in pre-pubertal and pubertal children (Carlson and Naughton, 1993; Buttifant et al., 1996; Naughton et al., 1997). Comparisons of AOD between studies have been viewed with caution in the past because of the variations in the methodology used (Bangsbo, 1996a). A comparison of the studies conducted by the aforementioned pediatric researchers (Carlson and Naughton, 1993; Buttifant et al., 1996; Naughton et al., 1997) is possible because the same methodology has been employed in the same laboratory. In general, the studies presented in Table 4.6 are in reasonably good agreement with each other. The data from the cycle ergometer study investigating AOD of pre-pubertal males conducted by Carlson and Naughton (1993) are in good agreement with those from the present study, despite a difference in the exercise-testing mode. AOD values of the pre-pubertal non-asthmatic males in the study conducted by Buttifant et al., (1996) are slightly larger, but within the S.E.M. for both studies, when compared with AOD values for the pre-pubertal boys in this study. AOD (ΣL) values from the pubertal male badminton players collected by Naughton et al., (1997) are much larger than those of the pubertal participants in the present study. At 120% of peak oxygen uptake the pubertal male participants in the study conducted by Naughton et al., (1997) produced AOD values of 4.3 L. These values are closer to those of the post-pubertal participants in the present study than to the pubertal participants. This may be explained by the observation by Naughton et al., (1997) that the participants had almost completed their pubertal development. The pubertal

participants in the present study were at a mean Tanner Stage of 3 for genital development, which represents mid-pubertal development.

When AOD values ($\Sigma mL.kg^{-1}$) were compared, the values of the pubertal participants in the study by Naughton et al., (1997) were only marginally higher than those of the pubertal participants in the present study. When compared with the post-pubertal group in the present study the pubertal group from the study conducted by Naughton et al., (1997) had AOD values that were almost 20 $mL.kg^{-1}$ higher. The participants in the study conducted by Naughton et al. (1997) were described as anaerobically well trained, as they were members of a state representative badminton squad. This may indicate that at this stage of pubertal development training could have an effect on the AOD.

Table 4.6 A comparison of accumulated oxygen deficit studies in children

Study	Subjects	(n)	Exercise Mode	Exercise Intensity	AOD (ΣL)	AOD ($\Sigma mL.kg^{-1}$)
Carlson & Naughton (1993)	Pre-pubertal males	9	Bicycle ergometer	110%	1.3	35.3
		9		130%	1.3	37.1
		9		150%	1.3	36.8
Buttifant et al., 1996	Pre-pubertal non-asthmatic males	10	Treadmill	110%	1.7	51.5
		10		130%	1.6	47.0
Naughton et al., 1997	Pubertal males	8	Treadmill	120%	4.3	71.5
		8		130%	4.1	67.6
Present study	Pre-pubertal males	10	Treadmill	120%	1.3	41.3
	Pubertal males	10			2.8	63.4
	Post-pubertal males	10			3.9	54.4

4.5.4 Aerobic and anaerobic contributions

The aerobic and anaerobic contributions to the supramaximal exercise bout can be estimated using the predicted total accumulated oxygen demand and the measure of accumulated oxygen uptake (Carlson & Naughton, 1998). Relative contributions can be determined by dividing the actual oxygen consumed during the test by the amount predicted for the exercise time. This calculation provides the percentage of the aerobic contribution and subsequent anaerobic contribution (Carlson and Naughton, 1998).

Medbø and Tabata (1989) investigated the relative importance of the aerobic and anaerobic contributions to the supramaximal test. The authors found that there was a considerable contribution from the aerobic energy sources even in supramaximal test times as low as 30 seconds. Supramaximal cycling tests of in varying durations of 30 seconds, 1 min and 2 minutes were conducted and aerobic contributions to these tests were 40%, 50%, and 65% of the total energy for the respective durations of 30, 60 and 120 seconds, respectively (Medbø and Tabata, 1989). Medbø (1991) therefore suggested that a 50/50 split of aerobic to anaerobic energy contributions would occur after a 1-minute high-intensity exercise bout and that the relative contribution of anaerobic energy sources diminished with increasing supramaximal test time.

The relative aerobic and anaerobic contributions to the supramaximal test in the present study (Figure 4.3) show a marked difference in the percentage contributions when the groups were compared. According to Medbø (1991), supramaximal test time will impact on the relative contribution of the aerobic and anaerobic energy systems to the supramaximal test. There were no differences in the supramaximal test times between the groups in this study. The post-pubertal group exhibited a 10% and 6% greater contribution from anaerobic energy sources, respectively when compared with the pre-pubertal and pubertal groups.

The aerobic contribution to the Wingate Anaerobic Test was examined in 101 boys who were aged between seven and fifteen years (Van Praagh *et al.*, 1989). When oxygen uptake during the test was expressed relative to body mass as well as the total work performed, there was a marked decrease in the aerobic contribution to the Wingate Anaerobic Test with increasing age. The findings of this study suggest a higher aerobic contribution and lower anaerobic contribution to a 30-second all-out test in the younger boys when compared with the older boys (Van Praagh et al., 1989). The findings of Van Praagh et al. (1989) show the same trend as the findings of the present study – the post-pubertal group exhibited a greater anaerobic contribution to the supramaximal test than either the pre-pubertal or pubertal groups. Studies investigating the muscle metabolism of children using ^{31}P NMRS suggested that pre-pubertal children are less able to generate ATP

through anaerobic pathways during high intensity exercise when compared with adults (Zanconato et al., 1993). Kuno et al. (1995) found that both trained and untrained 12 – 15 year old males also exhibited a lower anaerobic metabolism during exercise when compared with the adult subjects in the same study. A recent review of the anaerobic function of children (Dore *et al.*, 2000) supported the findings of Zanconato et al., (1993) and Kuno et al., (1995) of a lower anaerobic metabolism in children when compared with adults. Van Praagh (2000) suggested that hormonal changes and improved motor coordination might be the most important contributors to improved anaerobic performance that is observed during growth. These findings may explain the smaller anaerobic contribution of the pre-pubertal group to the supramaximal test when compared with both the pubertal and post-pubertal groups in this study.

4.5.5 Blood parameters

In order to provide more information about the anaerobic performances of the pre-pubertal, pubertal and post-pubertal participants in the present study, blood samples were taken from the participants prior to and post supramaximal exercise. The blood samples provided by the participants were analysed for lactic acid accumulation and changes in the pH level of the blood. Naughton et al., (1997) identified the measurement of lactic acid accumulation and changes in the pH level of the blood as useful in providing information about the relative contribution and capacity of the anaerobic pathways during high-intensity exercise tests using AOD in pubertal participants.

4.5.5.1 Acid/Base balance

There was a difference in the post-exercise pH concentrations when the pre-pubertal group was compared with the post-pubertal group only. There were no differences in the post-exercise pH values when the pubertal and post-pubertal groups were compared (Figure 4.6). All of the groups in the present study experienced a decrease in pH which indicates that there has been an increase in anaerobic metabolism (Åstrand *et al.*, 1986). There was a greater decrease in pH in the post-pubertal group when compared with the pre-pubertal and pubertal groups. Hebestreit et al., (1996) observed that the children in their study exhibited a faster time course for recovery of post-exercise acidosis than the adults in the same study. These findings support the faster recovery in post-exercise pH in the pre-pubertal participants when compared with the post-pubertal participants in this study. The greater decrease in the pH of the post-pubertal group is supported by the findings of Matejková et al., (1980). The authors investigated the pH response to maximal exercise in males (n = 103) aged 12 to 15 years in a study conducted over four years. An age

related decrease in the pH response to maximal exercise was observed. The decrease in pH is related to the increase in anaerobic metabolism; therefore, smaller decreases in pH as a result of high-intensity exercise may indicate differences in anaerobic metabolism in pre-pubertal and pubertal participants when compared with post-pubertal participants. The findings of the present study are supported by comparisons with adult data that are presented in Table 4.7 below. The table presents some results from adult studies that have measured blood pH after supramaximal exercise. These studies have used AOD as the means of evaluating anaerobic performance in pubertal and adult populations using treadmill running as the mode of exercise.

The comparisons in Table 4.7 support the notion that there may be a developmental decrease in blood pH after supramaximal exercise in adults when compared with that of children. The pre-pubertal and pubertal participants in the present study had a higher blood pH after supramaximal exercise when compared with that of all of the adults in the studies examined. The post-pubertal group exhibited a blood pH that was similar to the endurance runners in the study conducted by Hermanson and Medbø (1984), and to the recreational runners in the study conducted by (Olesen *et al.*, 1994).

Table 4.7 pH after supramaximal treadmill exercise in male adults and children

Authors	Subjects	(n)	pH
Hermansen and Medbø, 1984	Endurance runners	6	7.17
	Sprint runners	6	7.06
Olesen et al., 1994	Adult recreational runners	6	7.13
	Adult sprint & distance runners	8	6.95
Naughton et al., 1997	Pubertal badminton players	8	7.10
Present study	Pre-pubertal	5	7.22
	Pubertal	5	7.22
	Post-pubertal	5	7.17

4.5.5.2 Blood lactate

The post exercise blood lactate concentration of the post-pubertal group was greater than that of both the pre-pubertal and pubertal groups between 3 – 7 minutes post supramaximal exercise and again at 20 minutes post-exercise. The pubertal group had a significantly lower post-exercise blood lactate when compared with the post-pubertal group at 3 minutes post-exercise only (Figure 4.7). Comparisons between the post-exercise blood lactate concentrations of adults and children taken from AOD studies indicate that the adults have higher blood lactate concentrations post-exercise. Table 4.8 below illustrates these differences. All of the adult studies show a much higher blood lactate concentration when compared with that of the children in the present study. No other child-based studies AOD studies have reported blood lactate concentrations post-supramaximal exercise.

Table 4.8 Blood lactate responses after supramaximal treadmill exercise in male adults and children

Authors	Subjects	(n)	Mean Blood Lactate
Hermansen and Medbø, 1984	Adult endurance runners	6	12.6
	Adult sprint runners	6	17.0
Bangsbo et al., 1993	Adult soccer players	15	12.9
	Adult runners	14	10.7
Olesen et al., 1994	Adult recreational runners	6	10.4
	Adult sprint & distance runners	8	12.8
Present study	Pre-pubertal	5	3.77
	Pubertal	5	4.50
	Post-pubertal	5	7.20

4.5.5.3 The significance of haemoglobin and myoglobin in AOD

The ability of the body to transport and store oxygen has an impact on AOD. Medbø (1991) attributes a 10% error in AOD to the transport and storage of oxygen in haemoglobin and myoglobin assuming that the total muscle mass of the body is 25% of the body mass. The concentration of haemoglobin was of interest in this study because it is an indicator of the body's ability to transport oxygen in the blood. The haemoglobin concentration of the pre-pubertal participants was significantly lower than that of the pubertal and post-pubertal groups. The lower haemoglobin concentration observed in the pre-pubertal group suggests that they were not able to transport as much oxygen in the blood when compared with the pubertal and post-pubertal participants.

Oxygen is stored in the muscle as myoglobin. Myoglobin storage capacity is higher in type I fibres than all of the other fibre types (Åstrand and Rodahl, 1986). Previous research indicates that pre-pubertal children have a higher percentage of type I fibres (Bell *et al.*, 1980; Oertel, 1988; Lexell *et al.*, 1992). This suggests that the ability to store oxygen in the muscles may be greater in pre-pubertal children than in pubertal and post-pubertal children.

The figures reported by Medbø (1991) of a 10% error in AOD attributed to the storage of oxygen in the blood and muscles have been suggested for the adult population. Given the possible differences in the oxygen transport and oxygen storage capacity of children when compared with adults, we are unable to definitively suggest whether the error in AOD suggested by Medbø (1991) may be the same in children. We can only suggest that there may be differences.

This study found that there was an increase in AOD (ΣL) when the pre-pubertal group was compared with the pubertal and post-pubertal groups. The pre-pubertal group also contributed a higher percentage of energy aerobically when compared with the other groups. These findings are supported by smaller post-exercise indices of anaerobic energy production, blood lactate and pH in the pre-pubertal group. The results are in agreement with findings of a smaller anaerobic performance in less mature children when compared with more mature children in tests of anaerobic power.

Chapter 5

STUDY TWO

ASSESSMENT OF ACCUMULATED OXYGEN DEFICIT: A COMPARISON OF OXYGEN UPTAKE UTILISING DISCRETE AND CONTINUOUS INCREMENTAL SUBMAXIMAL PROTOCOLS

This study has been divided into two parts. The first part, Part 2A, deals with the comparison of oxygen uptake utilising discrete and continuous incremental submaximal protocols. The second part, Part 2B, deals with the repeatability of continuous incremental submaximal protocols.

PART 2A

OXYGEN UPTAKE UTILISING DISCRETE AND CONTINUOUS INCREMENTAL SUBMAXIMAL PROTOCOLS

Abstract

The study investigated the oxygen uptake in pre-pubertal males (n=10) using discrete and continuous incremental submaximal protocols. Discrete submaximal protocols (DSP) were defined as individual submaximal exercise bouts of four minutes duration with a recovery interval of at least 20 minutes between each exercise bout. No more than three discrete submaximal exercise bouts were conducted in one day. The continuous incremental submaximal protocol (CISP) required the participants to run on the treadmill for twelve minutes in total, with an incremental increase in treadmill speed every three minutes. Both the DSP and CISP were conducted at identical speeds of 6.0, 7.0, 8.0 and 8.5 $km.hr^{-1}$. Mean steady state oxygen uptake for each of the speeds was 31.26 ± 0.76, 38.84 ± 1.00, 42.59 ± 1.87, 47.90 ± 1.06 for DSP and 31.11 ± 0.96, 40.37 ± 1.47, 47.06 ± 1.24, 51.17 ± 1.18 for CISP, respectively. After the DSP and CISP had been conducted, individual linear regression equations were constructed to describe the relationship between submaximal oxygen uptake and exercise intensity for both the DSP [1.27 + 0.14(x)] and CISP [1.92 + 0.12(x)]. Individual linear regression equations were used to predict a supramaximal treadmill running speed that equated to a value representing a "theoretical"120% of the participant's peak oxygen uptake. The participants ran at the predicted speeds on the treadmill and expired gases were collected during each of the supramaximal tests. Actual oxygen uptake was measured for each of the tests and subtracted from the theoretical oxygen uptake for the duration of the supramaximal test. This was accumulated oxygen deficit. One-way analysis of variance (ANOVA) was used to determine whether there was a difference in oxygen uptake, individual regression line parameters, supramaximal treadmill speeds (DSP mean speed 11.78 ± 0.55 $km.hr^{-1}$ and CISP speed 12.63 ± 0.44 $km.hr^{-1}$)and AOD (DSP 1.14 ± 2.44 (ΣL), 45.35 ± 4.73 ($\Sigma mL.kg^{-1}$); CISP 0.90 ± 0.12 (ΣL), 35.73 ± 4.30 ($\Sigma mL.kg^{-1}$), between the DPS and CISP. Results of the ANOVA revealed no differences in any of the parameters when the DSP and CISP were compared. As there was no difference in the oxygen uptake between the DSP and CISP, these results suggest that either DSP or CISP may be used to determine the relationship between submaximal oxygen uptake and exercise intensity.

PART 2B

THE REPEATABILITY OF OXYGEN UPTAKE DURING CONTINUOUS INCREMENTAL SUBMAXIMAL PROTOCOLS

Abstract

The day-to-day measurement of oxygen uptake in children during submaximal tests has been shown to be reliable. These submaximal oxygen uptake tests have been discrete in nature. To date there have been no investigations into the day-to-day repeatability of oxygen uptake during continuous incremental submaximal protocols (CISP). Consequently, the purpose of this study was to examine the day-to-day repeatability of submaximal oxygen uptake during CISP on either the treadmill or cycle ergometer. Ten pre-pubertal participants who volunteered to participate in this study attended the laboratory at the same time on two occasions separated by one day. During their visit to the laboratory, the participants completed a CISP on each day in order to examine whether the oxygen uptake during the tests was repeatable. Paired t-tests were used to determine whether there were any differences in the oxygen uptake of the participants over the testing period. Paired samples correlations were calculated to investigate the repeatability of the oxygen uptake between the two CISP. The paired t-tests revealed that the oxygen uptake between the two CISP was not different. The paired samples correlations indicated that there was a strong relationship between the oxygen uptake during the CISP over three days.

5.1 Introduction

Determining the accumulated oxygen deficit (AOD) relies on establishing the relationship between oxygen uptake and exercise intensity at submaximal workloads. Medbø et al. (1988) describe a methodology to establish this relationship, which requires the subject to complete between 10 – 35 discrete submaximal exercise bouts. The relationship between oxygen uptake and exercise intensity during the discrete submaximal exercise bouts is then found using an individual linear regression equation. The y-intercept and the slope of the regression line are then used to predict a supramaximal workload for the subject. A supramaximal exercise test is then conducted. During the supramaximal exercise test expired gases are collected. Oxygen uptake during the supramaximal test is then calculated and is subtracted from the predicated (theoretical) oxygen uptake test, which is then the calculated AOD.

Determining oxygen uptake during submaximal exercise is an important component of determining AOD. At present, a number of investigators in this area (Withers et al., 1993; Tabata et al., 1996; Buck and McNaughton, 1999a; Weber and Schnider, 2000) utilise the submaximal methodology described by Medbø et al (1988) which is extremely time consuming. Reliable alternative methods of determining the relationship between oxygen uptake and submaximal exercise need to be examined. Continuous incremental submaximal protocols (CISP) offer a potential alternative, but other factors such as oxygen drift, an increase in oxygen uptake without a concomitant increase in exercise intensity, may make the determination of the relationship difficult at higher submaximal intensities. In order to determine whether oxygen drift would preclude the use of CISP, the impact of oxygen drift through an examination of the kinetics of oxygen uptake should be discussed.

5.1.1 The kinetics of oxygen uptake

The kinetics of oxygen uptake during submaximal constant load exercise is generally well described in adults (Whipp & Wasserman, 1972; Hagberg *et al.*, 1978; Barstow *et al.*, 1994) as well as children (Freedson *et al.*, 1981; Cooper *et al.*, 1984; Cooper, 1989; Zanconato *et al.*, 1991). The kinetics of oxygen uptake during constant-load submaximal exercise can be characterised by three phases. Phase one is characterised by a rapid increase in oxygen uptake after the onset of exercise; this phase usually lasts approximately 15 - 25 seconds. Phase two is characterised by a more rapid rise in oxygen uptake until a plateau is reached (Xu & Rhodes, 1999). This plateau is indicative of a steady state. Steady state represents the equilibrium between the oxygen cost of exercise and the oxygen supply to the exercising muscles (Robergs & Roberts, 1997). Oxygen uptake should remain constant for the duration of the submaximal exercise bout

once steady state has been reached, if the exercise intensity remains the same (Xu & Rhodes, 1999). Oxygen uptake during submaximal exercise of a long duration (60 minutes) (Asano & Hirakoba, 1984); (Rowland & Rimany, 1995) or high intensity (above anaerobic threshold) (Xu & Rhodes, 1999) has been shown to increase without a concomitant increase in exercise intensity. This increase in oxygen uptake, which is characterised as phase three, is termed the "slow component" of oxygen uptake, or oxygen drift.

5.1.1.1 The oxygen uptake kinetics of children

Several authors have described the oxygen uptake kinetics of children using submaximal (Cooper *et al.*, 1985; Armon *et al.*, 1991; Zanconato *et al.*, 1991) and maximal (Cooper *et al.*, 1984) exercise protocols. Studies investigating the oxygen uptake kinetics at submaximal intensities revealed no differences in the kinetics of adults when compared with those of children (Armon et al., 1991; Zanconato et al., 1991), and younger children (7 – 9 years) when compared with older children (15 – 18 years) (Cooper et al., 1985). Contrasting results have been found when the oxygen uptake kinetics of adults and children has been investigated during high intensity exercise. Some studies (Macek and Vavra, 1980; Sady, 1981; Armon et al., 1991) have found that oxygen uptake kinetics of children is different from those of adults. Armon et al. (1991) commented that the pattern of the oxygen uptake kinetics in children was "quantitatively and qualitatively different from that in adults" (Armon et al., 1991, page 845). The authors found that children adjusted to the exercise intensity at a faster rate (faster kinetics), and exhibited little or no oxygen drift and, if there was a drift, it was smaller than that of the adults studied. Children, however, had a greater oxygen cost of exercise when compared with the adults in the study (Armon et al., 1991). In contrast, Zanconato et al. (1991), found no differences in the oxygen uptake kinetics during high intensity exercise when children and adults were compared.

To date, no research has been conducted on the oxygen uptake kinetics of continuous incremental submaximal exercise in children.

5.1.2 Submaximal exercise protocols and accumulated oxygen deficit

Green and Dawson (1996a) examined the difference in oxygen uptake between DSP and CISP in a study examining accumulated oxygen deficit of adult cyclists. The authors found no differences in the oxygen uptake when the DSP and CISP were compared. Green and Dawson (1996a) were cautious about the possibility of oxygen drift (phase three) of oxygen uptake kinetics affecting the oxygen uptake, but found only evidence of an oxygen drift at the highest submaximal intensities during the CISP. As oxygen drift has been shown to be smaller in children than in adults (Armon

et al., 1991) a comparison of the oxygen uptake between discrete and continuous incremental submaximal exercise may be different in children. Alternative submaximal protocols, such as a CISP would also need to be reliable.

5.1.3 Repeatability of oxygen uptake in the pediatric population

Oxygen uptake has been shown to be reliable in submaximal treadmill running tests conducted on two different testing days (Unnithan, 1993; Rogers *et al.*, 1994; Frost *et al.*, 1995; Unnithan *et al.*, 1995). The studies (Unnithan 1993; Rogers et al. 1994; Frost et al. 1995; Unnithan et al., 1995) investigated the repeatability of oxygen uptake in submaximal exercise tests that permitted a rest period before the next submaximal exercise test was conducted. To date, there have been no studies conducted on the day-to-day repeatability of CISP.

5.1.4 Repeatability of discrete submaximal oxygen uptake in the pediatric population

In the past, oxygen uptake determined during a series of discrete submaximal exercise tests, with a rest interval between each submaximal exercise test has been used to determine the relationship between oxygen uptake and submaximal exercise intensity (Carlson & Naughton, 1993; Buttifant *et al.*, 1996; Carlson & Naughton, 1998). A large variation in the day-to-day oxygen uptake of children has been observed when discrete submaximal tests have been used to determine the relationship between submaximal oxygen uptake and exercise intensity. This variation in oxygen uptake with discrete submaximal exercise tests has caused some difficulties when using the data points (oxygen uptake and submaximal exercise intensity) to construct a linear regression that describes the relationship between oxygen uptake and submaximal exercise intensity. Previously, (Medbø, 1991) advocated the exclusion of data points that did not appear to be linear prior to the calculation of the linear regression equation. This method advocated by Medbø (1991) is problematic in children as the submaximal work capacity of children is smaller than that of adults and relatively fewer data points are available to describe the relationship between oxygen uptake and submaximal exercise intensity (Naughton and Carlson, 1998).

5.1.5 Continuous incremental submaximal protocols in adults

The use of a CISP to evaluate the relationship between oxygen uptake and submaximal exercise intensity for the determination of AOD has been used with adult populations only (Craig, et al. 1993; Green and Dawson, 1996a; Green and Dawson, 1996b). Green and Dawson (1996a) found no differences in the regression equations constructed to describe the relationship between oxygen uptake and submaximal exercise intensity after two continuous incremental tests were

conducted on two separate days. As these tests were conducted on an adult population, and the repeatability of oxygen uptake in adults have been shown to be different when compared with those of children (Freedson *et al.*, 1981; Sady *et al.*, 1989), examining the repeatability of oxygen uptake from CISP in children is important.

Carlson and Naughton (1998) suggested that investigating different protocols to determine the relationship between submaximal oxygen uptake and exercise intensity was a direction of future research in this area. The authors advocated that less time consuming, but reliable alternative protocols for determining the relationship between submaximal oxygen uptake and exercise intensity should to be developed for use with the pediatric population (Carlson & Naughton, 1998).

5.2 Purpose

The purpose of this study was twofold: the first to compare the oxygen uptake at various submaximal exercise intensities using discrete and continuous incremental submaximal protocols. The second purpose was to examine the repeatability of oxygen uptake during continuous incremental submaximal protocols.

5.3 Methodology

The methodology employed in this study is divided according to those used in Study 2A and Study 2B. Methods common to both studies, such as the determination of sexual maturation and statistical analysis of the data, are combined at the end of the methodology section.

The methodology employed during this study will be presented in 3 major sections: the participants and the testing timetable, oxygen uptake measures, discrete and continuous incremental submaximal protocols.

5.3.1 The participants and the testing timetable

Ten pre-pubertal males volunteered to participate in this study. Prior to the commencement of this study, all participants came to the laboratory for a familiarisation session. The conduct of the familiarisation session has been described previously (Chapter 3). The participants attended the laboratory for testing on at least six occasions. Submaximal testing sessions Two to Four were randomised. The details of tests conducted in each session are presented in Table 5.1 below and in Chapter 3.

Table 5.1 Testing schedule in Study 2A

Session	Test Conducted
One	Peak oxygen uptake
Two - four	Two to three discrete submaximal oxygen uptake determinations Or Continuous incremental submaximal oxygen uptake determinations
Five - six	Supramaximal test with workload predicted from linear regressions conducted using either the discrete or the continuous submaximal workload/oxygen uptake relationships

5.3.2 Oxygen uptake measures

This study required the participants to run at identical submaximal running speeds using both DSP and CISP. The participants were required to reach steady state during each of the DSPs and before the next incremental load was applied during the CISP. Steady state oxygen uptake was defined as the point where there was equilibrium between the oxygen cost of exercise and the oxygen supply to the exercising muscles (Åstrand & Rodahl, 1986). This point was determined metabolically when the oxygen cost of exercise ($\dot{V}O_2$) reached a plateau after the initial onset of exercise. For the purposes of this study, steady state was achieved when the difference in oxygen uptake ($\dot{V}O_2$) at each 15-second measure, during the final minute of the exercise bout was no more than ± 2 ml $kg^{-1}.min^{-1}$. Each submaximal exercise workload was three minutes in duration. Three-minute stages have been shown as sufficient to attain steady state oxygen uptake in children in the past (Rowland, 1993).

5.3.3 Discrete and continuous incremental submaximal protocols

5.3.3.1 Discrete submaximal protocol

Each participant participated in a range of between 4 and 6 DSPs. The treadmill speeds set for the DSPs were: 6.0, 7.0, 8.0 and 8.5 $km.hr^{-1}$. Treadmill speed was calibrated before each submaximal testing session. DSPs were conducted at submaximal intensities in the range that represented between 30 and 90% of the participant's predetermined peak oxygen uptake. No more than three

discrete submaximal exercise bouts were conducted in one day. The participants also had a recovery interval of at least 20 minutes between each discrete determination.

5.3.3.2 Continuous incremental submaximal protocol

In order to determine whether there were differences in oxygen uptake when compared with the DSP, a CISP was also used. Each participant completed at least four workload increments in the CISP. The CISP were conducted at identical submaximal treadmill speeds (6.0, 7.0, 8.0 and 8.5 $km.hr^{-1}$) to the DSP, which also represented a range of between 30 and 90% of the participant's predetermined peak oxygen uptake.

5.3.4 Study 2B

The methodology employed during this section of the study will be presented in two sections: the participants and continuous incremental exercise protocols.

5.3.5 The participants

Ten pre-pubertal males volunteered to participate in this study. All participants attended a familiarization session as described previously (Chapter 3). The participants attended the laboratory on two different occasions. The testing was conducted over three days with a rest day separating each session.

5.3.6 Continuous incremental submaximal protocols

CISPs were conducted on a motorised treadmill or a cycle ergometer. Each participant completed at least four workload increments during the CISP on either the treadmill or the cycle ergometer. Each submaximal workload was between three and four minutes in duration. The participants were required to reach steady state, as defined previously. At the first testing session the participant completed a CISP on either a motorised treadmill or a cycle ergometer. Care was taken to ensure that both testing sessions were conducted at the same time of day to decrease the possible impact of diurnal variation on oxygen uptake. The second testing session was conducted in the same manner as the first. Two of the participants completed tests on both the cycle ergometer and the treadmill. The cycle ergometer and treadmill were used to investigate repeatability of both modes for investigations in Study 3.

5.3.7 *Determination of sexual maturation*

During the testing period, each participant's parents were asked to identify their child's level of sexual maturity according to the Tanner Scale (Tanner, 1962) (Appendix 2.2).

5.3.8 *Statistical analysis of the results*

The descriptive data was expressed as means ± standard errors (Mean ± S.E.M.). Linear regression equations were constructed to describe the relationship between submaximal oxygen uptake and exercise intensity in both sections of the study. In study 2A one-way analysis of variance (ANOVA) with repeated measures was used to determine the difference in the submaximal oxygen uptake when the DSP and CISP were compared. ANOVA was also used to locate differences in the slope and intercept of the linear regression, the supramaximal workloads and AOD.

In study 2B, paired t-tests were used to determine if there were any differences in the oxygen uptake of the participants over the testing period. Paired samples correlations were calculated to investigate the degreerepeatability of the relationship of oxygen uptake between the two CISPs. An alpha level of 0.05 was adopted for all testing.

5.4 Results

The focus of this methodological research was to examine the differences between submaximal oxygen uptakes measured at identical treadmill speeds using DSP and CISP and to determine the repeatability of oxygen uptake during CISP. The results of this study will be presented in two major sections, with subsections for studies 2A and 2B. Each subsection will present participant profiles, the oxygen uptake measures associated with submaximal protocols, and linear regression equations for submaximal protocols. Accumulated oxygen deficit measures will be presented for study 2A only.

5.4.1 Participant Profiles

5.4.1.1 Descriptive characteristics

The descriptive characteristics of the participants (n = 20) are presented in Table 5.2. The participants in study 2A were heavier (90^{th} percentile) and taller (97^{th} percentile) than the average Caucasian male of the same age (NHANES, 1992) Participants in study 2B were of average mass (50^{th} percentile) and slightly taller (75^{th} percentile) than the average Caucasian male of the same age (NHANES, 1992).

Table 5.2 Descriptive characteristics

Study section	Age (yr)	Mass (kg)	Height (cm)
2A	10.2	40.36	152
	± 0.33	± 2.18	± 1
2B	11.5	39.80	150
	± 0.43	± 2.04	± 2

Mean ± S.E.M.

5.4.1.2 Maximal effort profiles

The maximal effort profiles of the participants who participated in study 2A are presented in Table 5.3. Participants in study 2B did not undergo a peak oxygen uptake test.

The absolute peak oxygen uptake ($L.min^{-1}$) ranged between 0.99 - 2.11 $L.min^{-1}$. The relative peak oxygen uptake ($mL.kg^{-1}.min^{-1}$) of the participants ranged between 52.39 - 72.22 $mL.kg^{-1}.min^{-1}$. All of the participants attained maximal performances by fulfilling at least one of the criteria suggested by (Zwiren, 1989). These criteria (Zwiren, 1989) include either a peak heart rate that was 95% of their age-predicted maximum, a respiratory exchange ratio (RER) in excess of 1.05, or a "plateau" of oxygen uptake with increasing workload (Zwiren, 1989). Fifty percent of the participants fulfilled the heart rate criteria while the other forty percent attained the RER criteria. Fifty percent of the participants also exhibited a plateau in oxygen uptake with increasing workload.

Table 5.3 Maximal effort profiles

Peak $\dot{V}O_2$ ($L.min^{-1}$)	Peak $\dot{V}O_2$ ($mL.kg^{-1}.min^{-1}$)	Peak RER	Peak heart rate ($b.min^{-1}$)
1.66	64.31	1.05	198
± 0.11	± 2.28	± 0.02	± 2

Mean ± S.E.M.

5.4.2 *Oxygen uptake measures during discrete and continuous incremental submaximal protocols*

Study 2A required the participants to run on a treadmill at speeds of 6.0, 7.0, 8.0, and 8.5 $km.hr^{-1}$ using both DSP and CISP. Table 5.4 presents the mean oxygen uptakes ($mL.kg^{-1}.min^{-1}$) at each of the identical submaximal speeds for the DSP and CISP. No differences in oxygen uptake were found between either of the DSP or CISP at any of the selected submaximal treadmill speeds ($p > 0.05$) (Table 5.4, Figure 5.1) (Appendix 5.1). There was, however, a trend for consistently higher oxygen uptake during the CISP when compared with the DSP. The trend represented a 3.8% larger oxygen uptake at 7.0 $km.hr^{-1}$, a 9.5% larger oxygen uptake at 8.0 $km.hr^{-1}$ and a 6.4% larger oxygen uptake at 8.5 $km.hr^{-1}$.

5.4.2.1 Oxygen uptake during continuous submaximal exercise

Participants in study 2B completed two separate testing sessions spread over three days. The oxygen uptake during the CISP is presented in Table 5.5 and Figure 5.2. There were no

differences in the oxygen uptake when the two testing sessions were compared ($p > 0.05$) (Appendix 5.2).

5.4.2.2 Repeatability of oxygen uptake during continuous incremental submaximal protocols

The repeatability of the oxygen uptake over the testing period was examined statistically using paired samples correlations. The paired samples were all strongly correlated with each other. Table 5.6 presents the paired samples correlations of the oxygen uptake during CISP (Appendix 5.2).

Table 5.4 Mean steady state oxygen uptake during DSP and CSIP

	$\dot{V}O_2$ (ml. kg^{-1}. min^{-1})			
Speed (km. hr^{-1})	**Discrete**	**Continuous**	**% difference**	**P value**
6.0	31.26 ± 0.76	31.11 ± 0.96	0.5	0.90
7.0	38.84 ± 1.00	40.37 ± 1.47	3.8	0.40
8.0	42.59 ± 1.87	47.06 ± 1.24	9.5	0.06
8.5	47.90 ± 1.06	51.17± 1.18	6.4	0.05

Mean ± SEM.

Table 5.5 A comparison of oxygen uptake during CSIP

Submaximal Workload	Oxygen Uptake ($mL.kg^{-1}.min^{-1}$) Testing Session One (day one)	Oxygen Uptake ($mL.kg^{-1}.min^{-1}$) Testing Session Two (day three)
One	22.55	22.46
	± 1.23	± 1.43
Two	30.33	30.83
	± 0.96	± 1.22
Three	35.93	36.54
	± 1.12	± 1.45

Mean ± S.E.M.

Table 5.6 Paired sample correlations of oxygen uptake during CSIP

Submaximal Workload	Correlation	Significance
One	0.933	0.000
Two	0.849	0.000
Three	0.885	0.000

5.4.2.3 Differences in RER and V_E l. min^{-1} in the final two minutes of the submaximal tests when the discrete and continuous incremental protocols were compared.

A comparison of the data from expired gases for the final two minutes of the three-minute workloads revealed differences in RER and expired ventilation (V_E l. min^{-1}) between DSP and CISP ($p < 0.05$). The mean RER was 5.7% higher in the final workload of the continuous submaximal exercise protocol when compared with the DSP. The mean V_E ($L.min^{-1}$) was 14.7% higher in the final workload of the continuous submaximal exercise protocol when compared with the DSP. Figures 5.3 and 5.4 illustrate these differences (Appendix 5.1). There were no differences ($p < 0.05$) in oxygen uptake, percentages of F_EO_2 and F_ECO_2 between the DSP and CISP at identical treadmill speeds.

5.4.2.4 Differences in heart rate in the final two minutes of the submaximal tests when the DSP and CSIP were compared.

Heart rate was recorded during each minute of exercise during both the DSP and CISP. There were no differences ($p< 0.05$) in the heart rate recorded during the first two submaximal workloads (6.0 $km.hr^{-1}$ and 7.0 $km.hr^{-1}$) between the DSP and CISP. There were differences ($p < 0.05$) in the heart rates recorded at each minute between the DSP and CISP during the final two workloads (8.0 $km.hr^{-1}$ and 8.5 $km.hr^{-1}$). These differences represented a 6.8% increase in the mean heart rate (across the three minute workload) recorded during the CISP when compared with the DSP at a treadmill speed of 8.0 $km.hr^{-1}$. The mean heart rate recorded during the final workload (8.5 $km.hr^{-1}$) was 10.9% higher during the CISP when compared with the DSP. There were no differences in the heart rates within the protocols during the final two workloads. The CISP exhibited a 4.4% higher mean heart rate values at 8.0 $km.hr^{-1}$ were compared with the mean heart rates obtained at 8.5 $km.hr^{-1}$. These differences are illustrated in Figure 5.5 (Appendix 5.1).

5.4.2.5 Mean heart rates during CSIP

Heart rate was also recorded during the each of the CISP in Study 2B. Table 5.7 presents the mean heart rate at each of the submaximal workloads. There was a difference ($p<0.05$) between the mean heart rates when the first submaximal workloads were compared. There were no differences ($p>0.05$) in the mean hearts rate when the subsequent three workloads were compared (Appendix 5.2).

A comparison of the mean heart rate during the first submaximal workload indicates that the mean heart rate was 4.1% greater on the second testing session (day 3) when compared with the first testing session (day 1). The percentage difference between the mean heart rates becomes progressively smaller as the submaximal workload increased in intensity. The percentage difference between the mean heart rates at workload two was 1.5% and workload three 1.3%. The mean heart rate was consistently higher during testing session two (day 3).

The repeatability of the mean heart rates over the testing period was examined statistically using paired samples correlations. The paired samples were all strongly correlated with each other (Appendix 5.2). Table 5.8 presents the paired samples correlations of the mean heart rate uptake during continuous incremental submaximal exercise.

Table 5.7 Comparison of mean heart rates during CSIP

Submaximal Workload	Mean Heart Rate ($b.min^{-1}$) Testing Session One (Day one)	Mean Heart Rate ($b.min^{-1}$) Testing Session Two (Day three)
One	119	124
	± 4	± 3
Two	140	142
	± 4	± 2
Three	156	158
	± 4	± 3

Mean ± S.E.M.

Table 5.8 Paired sample correlations of mean heart rate during CISP

Submaximal Workload	Correlation	Significance
One	0.755	0.005
Two	0.859	0.000
Three	0.905	0.000

5.4.2.6 Within-participant variation in oxygen uptake and heart rate during CSIP

The within-participant variation in oxygen uptake and heart rate during CISP was calculated by computing the difference between the participant's scores from test 1 and test 2, and divided by the mean of the scores from test 1 and test 2. This value represented the individual percentage variation between test 1 and test 2 (Rogers *et al.*, 1994). The individual values were then averaged to express the mean within-participant variation in oxygen uptake and heart rate between the two separate testing sessions over three days.

5.4.2.7 Mean within-participant variation in oxygen uptake during CSIP

The mean and range of within-participant variation in oxygen uptake during CISP is presented in Table 5.9. The greatest mean within-participant variation in oxygen uptake is observed at the second submaximal intensity. The within-participant variation at the second submaximal intensity

is 3.8% greater and 5% greater than the first submaximal intensity and the third submaximal intensity, respectively.

Table 5.9 Mean and range of within-participant variation for oxygen uptake during CSIP

Submaximal Workload	Mean within-participant variation	Range within-participant variation
One	5.67 ± 1.77	0.80 - 19.70
Two	5.89 ± 1.24	0.40 - 14.00
Three	5.60 ± 1.06	0.10 - 10.30

Mean ± S.E.M.

5.4.2.8 Mean within-participant variation in heart rate during CSIP

The mean and range of within-participant variation of heart rate during CISP is presented in Table 5.10. The greatest mean within-participant variation in heart rate is observed at the first submaximal intensity. The within-participant variation at the first submaximal intensity is 15.4% greater and 40.6% greater than the second submaximal intensity and the third submaximal intensity, respectively. The second submaximal intensity is 30.5% greater than the third submaximal intensity.

5.4.2.9 The coefficient of variation in oxygen uptake and heart rate during CSIP

The coefficient of variation was calculated for oxygen uptake and heart rate for the two testing sessions over three days. The coefficient of variation (CV) was calculated by dividing the standard deviation (SD) by the mean (m) and multiplying by 100 (CV = (SD/m) x 100). The coefficient of variation represents the sample variation relative to the sample mean (Armstrong & Costill, 1985).

Table 5.10 Mean and range of within-participant variability in heart rate during CSIP

Submaximal Workload	Mean within-participant variation	Range within-participant variation
One	6.53 ±1.62	1.60 - 18.00
Two	5.58 ± 0.85	1.40 - 8.90
Three	3.88 ± 0.80	0.60 - 9.20

Mean ± S.E.M.

5.4.2.10 The coefficient of variation in oxygen uptake during CSIP

The intraindividual variability in oxygen uptake during CISP conducted during two separate testing sessions over three days is presented in Table 5.11. The intraindividual variability in oxygen uptake was greater in the second testing session when compared with that of the first testing session. The intraindividual variability in oxygen uptake was 21.2% greater in workload one, 19.2% greater in workload two and 21.2% greater in workload three.

5.4.2.11 The coefficient of variation in the heart rate during CSIP

The intraindividual variability in heart rate during CISP conducted during two separate testing sessions over three days is presented in Table 5.12. The intraindividual variability in heart rate was greater in the first testing session when compared with the second testing session. The intraindividual variability in heart rate was 18.2% greater in workload one, 41.8% greater in workload two and 32.9% greater in workload three.

Table 5.11 Coefficient of variation in oxygen uptake during CSIP

Submaximal Workload	Testing Session One (day one)	Testing Session Two (day three)
One	18.6	22.0
Two	11.0	13.6
Three	10.8	13.7

Table 5.12 Coefficient of variation in heart rate during CSIP

Submaximal Workload	Testing Session One (day one)	Testing Session Two (day three)
One	11.6	9.5
Two	10.8	6.4
Three	10.5	8.1

5.4.2.12 The intensity of submaximal exercise

Medbø(Medbø & Tabata, 1989) and Tabata (1989) concluded that steady state submaximal exercise should be conducted at intensities representing 30 to 90% of $\dot{V}O_2$ peak. Table 5.13 presents the mean and range of percentage of $\dot{V}O_2$ peak that occurred at each submaximal speed and between the DSP and CISP.

No differences were reported between the percentages of peak $\dot{V}O_2$ when the DSP and CISP were compared ($p > 0.05$) (Appendix 5.3). There was, however, a consistent trend in both the mean (%) peak $\dot{V}O_2$ and range (%) peak $\dot{V}O_2$ to be higher during CISP when compared with DSP. When compared, the percentage differences represented a 3.9%, 9.5% and 5.8% greater oxygen uptake during CISP compared with DSP. The range (%) peak $\dot{V}O_2$ also showed a higher percentage difference when CISP was compared with DSP at the highest submaximal intensities (8.0 and 8.5 $km.hr^{-1}$). For the low end of the range, the oxygen uptake for CISP was 9.5% and 5.8% greater than that of DSP. At the high end of the range, the oxygen uptake for CISP was 3.4%, 3.8%, 5.8% and 10.5% greater than the oxygen uptake for DSP at 6.0, 7.0, 8.0 and 8.5 $km.hr^{-1}$, respectively.

Results presented in Table 5.13 is within the range suggested by (Medbø & Tabata, 1989) as appropriate intensities for determining submaximal oxygen uptake.

Table 5.13 Mean and range of peak oxygen uptake (%) at each speed and between protocols

Speed (km. hr^{-1}.)	Protocol	Mean peak $\dot{V}O_2$	Range peak $\dot{V}O_2$
6.0	D		
	C	48.93 ± 1.45	40.30 - 54.10
7.0	D	60.81 ± 1.86	52.03 - 70.64
	C	63.22 ± 2.44	48.93 - 73.41
8.0	D	66.85 ± 2.86	53.05 - 79.52
	C	73.82 ± 2.63	59.91 - 84.40
8.5	D	72.83 ± 2.18	62.2 - 79.50
	C	77.25 ± 3.09	65.2 - 88.80

Mean ± SEM D denotes DSP; C denotes CISP

5.4.2.13 Learning adaptations to the skills of treadmill running

In order to examine the possible effects of a learning adaptation to the skills of treadmill running throughout the testing period, mean oxygen uptake and exercise heart rates at 6km.hr^{-1} were compared over three visits to the laboratory during the testing period. There were no differences in the mean oxygen uptake or the exercise heart rates at 6km.hr^{-1} across the testing period ($p > 0.05$) (Appendix 5.4). Table 5.14 presents the mean oxygen uptake and heart rate data recorded on three separate occasions during the testing period.

5.4.3 Linear regression equations

5.4.3.1 Study 2A

Individual linear regression equations were constructed to describe the relationship between submaximal oxygen uptake and exercise intensity for both the DSP and CISP. The linear regression equations were used to predict the treadmill speed representative of 120% of the

participant's peak oxygen uptake. The procedure for calculating individual linear regression equations has been discussed previously (Chapter 3) (Appendix 3). The mean linear regression equations constructed from submaximal oxygen uptake and exercise intensities using both DSP and CISP were compared. Table 5.15 (Appendix 5.5) presents the mean linear regression equations using data from DSP and CISP. No differences were reported in any of the parameters of the linear regression equation used to predict the supramaximal speed that represented 120% of peak oxygen uptake in the sample group.

The accountable variance (R^2) represents the variance in the confidence in the linearity of the regression line. The R^2 expresses the variance in the relationship between the submaximal oxygen uptake and the exercise intensity for DSP and CISP. There was an accountable variance of 96% for both DSP and CISP.

Table 5.14 Mean oxygen uptake and heart rate at 6 km.hr^{-1} over three testing sessions

Visit Number	Oxygen uptake (mL.kg^{-1}.min^{-1})	Heart rate (b.min^{-1})
Visit One	31.52 ± 0.71	138 ± 4
Visit Two	29.99 ± 1.15	129 ± 4
Visit Three	31.11 ± 0.96	136 ± 4

Mean ± S.E.M.

Individual linear regression relationships were used to predict supramaximal treadmill speeds that were representative of 120% of the participant's peak oxygen uptake. The range of these speeds was 11.15 - 15.13 km.hr^{-1} for DSP and 9.58 - 15.17 km.hr^{-1} for CISP. The predicted mean supramaximal treadmill speeds, for subsequent supramaximal testing to determine AOD, were 11.78 ± 0.55 km. hr^{-1} for DSP and 12.63 ± 0.44 km. hr^{-1} for CISP. The two speeds were not different ($p > 0.05$) from each other. The mean treadmill speed for the supramaximal test predicted from CISP was 6.8% greater than that predicted from DSP.

5.4.3.2 Linear regression equations in study 2B.

The linear regression equations that describe the relationship between submaximal oxygen uptake and exercise intensity for the two tests are illustrated in Figure 5.2. There was no difference ($p>0.05$) (Appendix 5.6) in the y-intercept or slope of the regression lines constructed to describe

the relationship between submaximal oxygen uptake and exercise intensity when the two separate CSIPs were compared. Both of the regression lines exhibited a strong Pearson correlation coefficient of r = 0.99.

Table 5.15 Mean linear regression equation

Parameter	Protocol	Regression Value	Alpha
Y-intercept			**0.153**
	D	1.27 ± 0.27	
	C	1.92 ± 0.33	
Slope			**0.054**
	D	0.14 ± 0.00	
	C	0.12 ± 0.00	
Pearson Correlation (r)			**0.531**
	D	0.98 ± 0.00	
	C	0.98 ± 0.00	
Accountable Variance (R^2)			
(R^2) x 100			
	D	96%	
	C	96%	

Mean ± SEM; D denotes DSP; C denotes CISP

5.4.4 Accumulated oxygen deficit measures

5.4.4.1 The supramaximal test

Two supramaximal tests (at 120% peak oxygen uptake) were conducted in a random order. One of the supramaximal tests was conducted at a treadmill speed (mean 11.78 ± 0.55 km. hr^{-1}) calculated using the linear relationship between submaximal oxygen uptake and exercise intensity from DSP. This supramaximal test will now be called the discrete supramaximal test (DST). The other supramaximal test was conducted at a treadmill speed (mean 12.63 ± 0.44 km. hr^{-1}) that was calculated using the linear relationship between the submaximal oxygen uptake and exercise intensity from CSIP. This supramaximal test will now be called the continuous supramaximal test (CST).

The maximal heart rate, RER and supramaximal test time for each of the DST and CST is presented in Table 5.16. No differences ($p > 0.05$) were reported in the maximal heart rate, RER or supramaximal test time when DSP was compared with CISP. The supramaximal test time for DST was 11.1% longer than that of CST. The RER value for the CST was 3.8% greater than that of the DST.

Table 5.16 Maximal heart rate, RER and supramaximal test time measured using discrete and continuous protocols

	Protocol	
Measure	**Discrete**	**Continuous**
Maximal Heart Rate ($b.min^{-1}$)	189 ± 2	190 ± 1
RER	1.02 ± 0	1.06 ± 0
Test Time (s)	128.12 ± 14.59	114.01 ± 13.15

Mean ± SEM

5.4.4.2 Accumulated oxygen deficit

AOD is the difference between the predicted (theoretical) oxygen demand and the actual oxygen uptake for the supramaximal exercise bout. AOD measures are presented in Table 5.17. No differences were reported in the absolute (ΣL) or relative ($\Sigma mL.kg^{-1}$) AOD as a result of the DST or CST ($p > 0.05$) (Appendix 5.7). The absolute AOD (ΣL) was 21.1% greater and the relative AOD ($\Sigma mL.kg^{-1}$) 21.3% greater when the DST was compared with the CST. The greater AOD observed with the DST might be related to the longer supramaximal test time (11.1%) when this test was compared with the CST.

Table 5.17 Accumulated oxygen deficits measured using discrete and continuous protocols

	Protocol	
Measure	**Discrete**	**Continuous**
AOD (ΣL)	1.14 ± 2.44	0.90 ± 0.12
AOD ($\Sigma mL.kg^{-1}$)	45.35 ± 4.73	35.73 ± 4.30

Mean ± S.E.M.

Figure 5.1 Mean steady state oxygen uptake

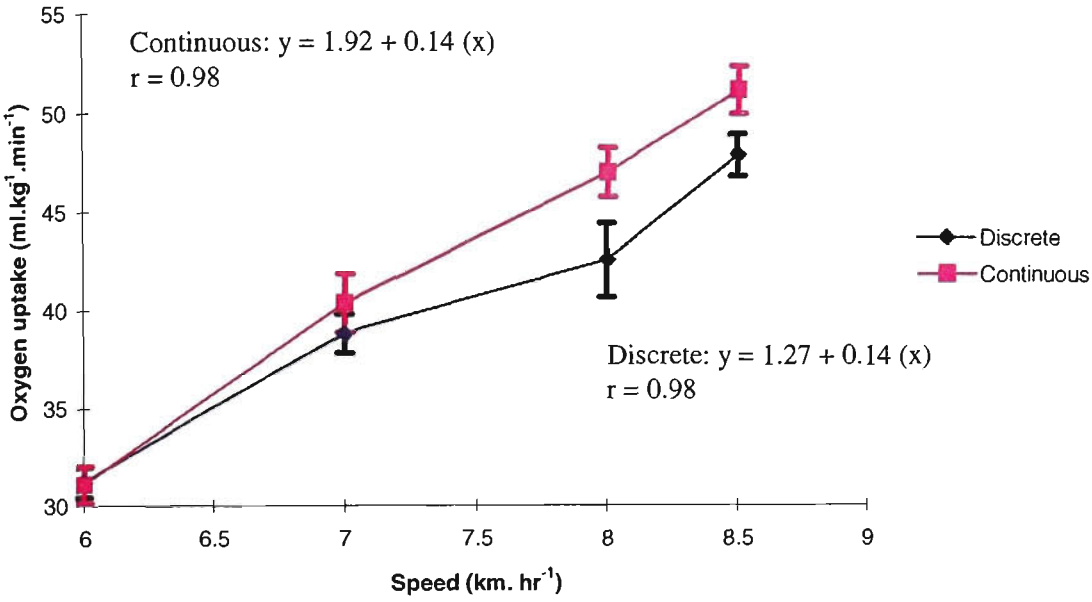


Figure 5.2 Comparison of oxygen uptake during CISP

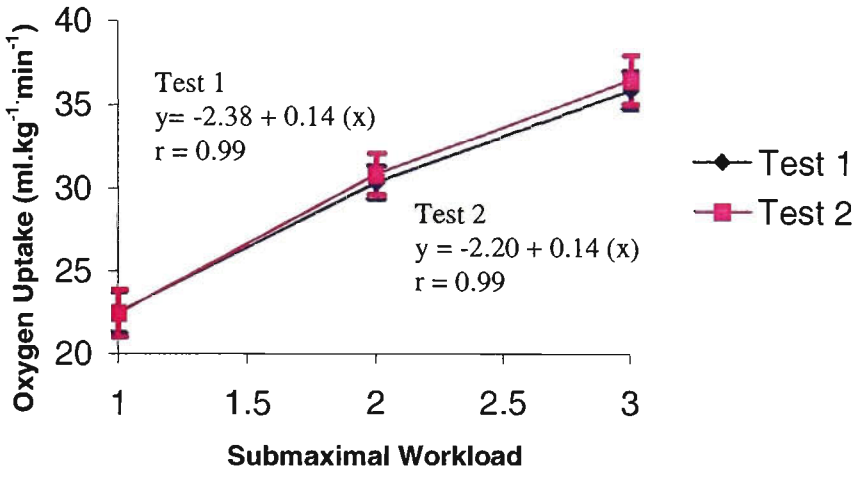
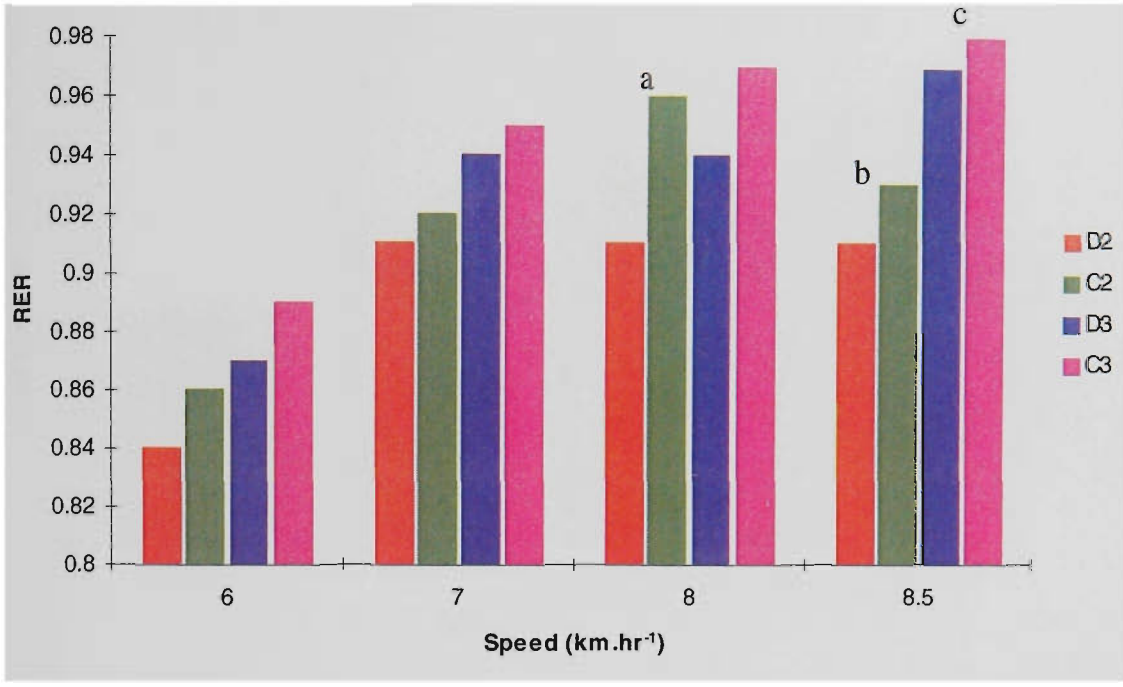


Figure 5.3 A comparison of RER in the last two minutes of exercise



D2 - discrete protocol min 2; C2 - continuous min 2; D3 - discrete protocol min 3;

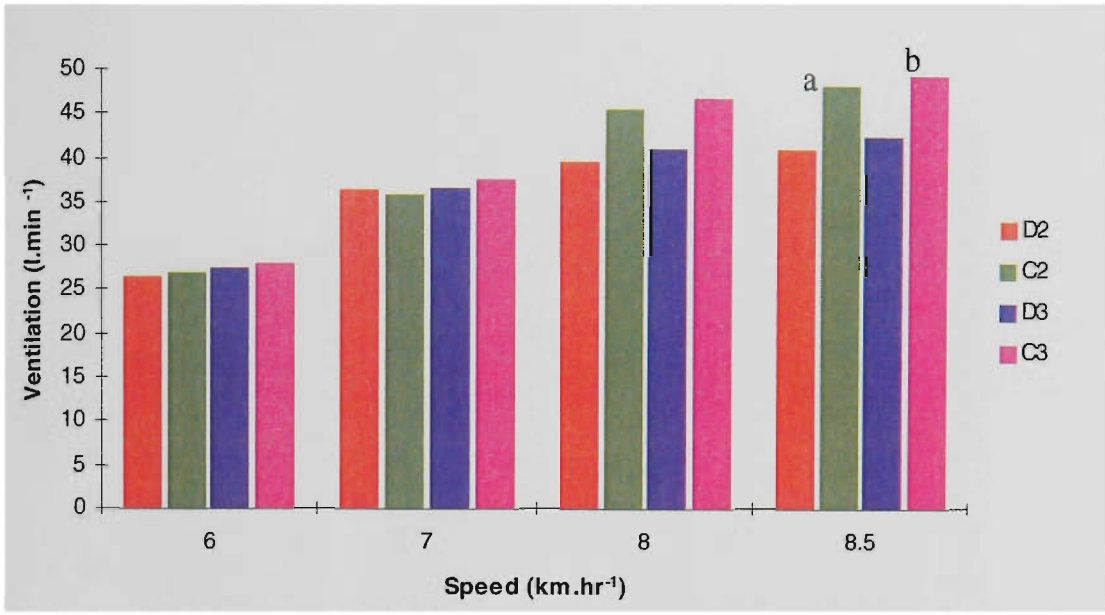
C3 - continuous protocol min 3. **$p > 0.05$**

'a' denotes continuous min 2 higher than discrete min 2 at 8.0 $km.hr^{-1}$

'b' denotes continuous min 2 higher than discrete min 2 at 8.5 $km.hr^{-1}$

'c' denotes continuous min 3 higher than discrete min 3 at 8.5 $km.hr^{-1}$

Figure 5.4 A comparison of ventilation in the last two minutes of exercise



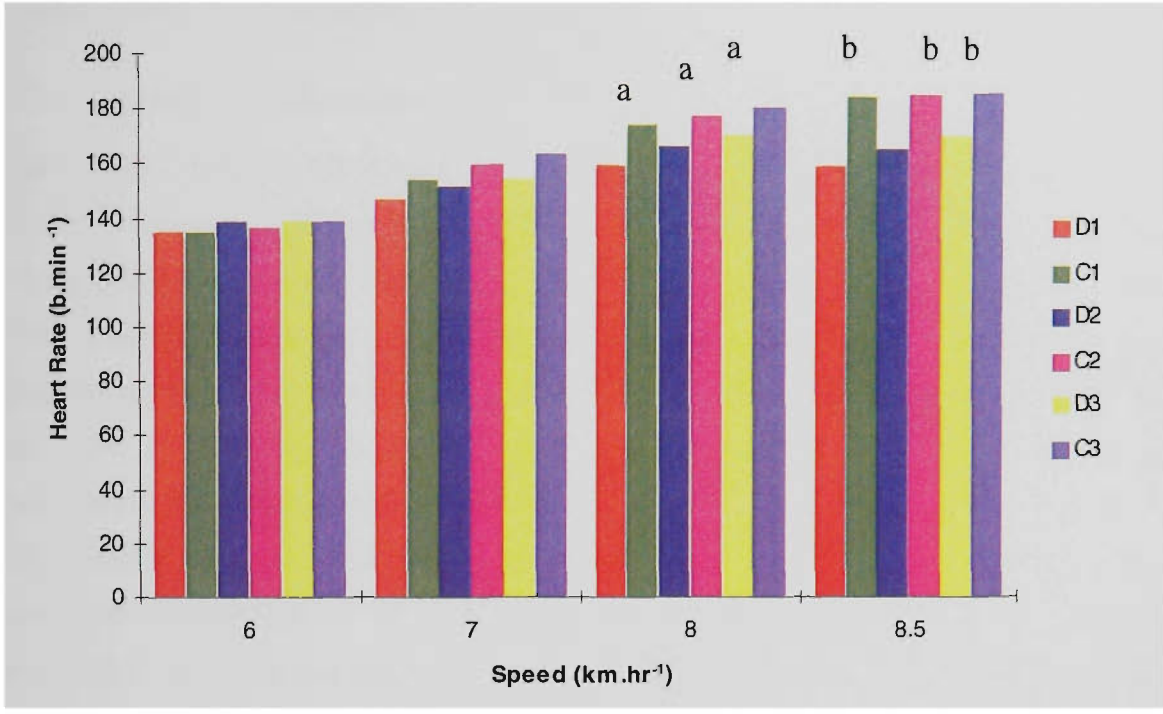
D2 - discrete protocol min 2; C2 - continuous min 2; D3 - discrete protocol min 3;

C3 - continuous protocol min 3. $p > 0.05$

'a' denotes continuous min 2 higher than discrete min 2 at 8.5 $km.hr^{-1}$

'b' denotes continuous min 3 higher than discrete min 3 at 8.5 $km.hr^{-1}$

Figure 5.5 A comparison of heart rate during the last two minutes of exercise



D1 - discrete protocol min 1; C1 - continuous protocol min 1; D2 - discrete protocol min 2; C2 - continuous min 2; D3 - discrete protocol min 3; C3 - continuous protocol min 3.

'a' denotes continuous protocol heart rates higher than discrete protocol heart rates at 8.0 km.hr-1 ($p > 0.05$)

'b' denotes continuous protocol heart rates higher than discrete protocol heart rates at 8.5 km.hr-1 ($p > 0.05$)

5.5 Discussion

This study investigated the oxygen uptake at identical submaximal intensities using DSP and CISP and the repeatability of oxygen uptake during CISP. No differences were reported between the protocols in this sample of pre-pubertal males. There was no difference ($p > 0.05$) in the oxygen uptake between the two testing sessions.

5.5.1 Oxygen uptake measures

Although not different, the oxygen uptake was consistently higher when CISP was compared with DSP. The observed differences in oxygen uptake during CISP has previously been described as "aerobic drift" (Rowland, 1996) or "oxygen drift" (Dick & Cavanagh, 1987; Westerlind *et al.*, 1992; Robergs & Roberts, 1997). Rowland (1996) describes "aerobic drift" as the slow rise in oxygen uptake that is observed during prolonged submaximal exercise. "Oxygen drift" was described by (Robergs & Roberts, 1997) as an increase in oxygen consumption during presumably steady state exercise when the exercise has been performed for an extended period of time. Steady state, for the purposes of this study, was denoted as a difference in the oxygen uptake of no more than ± 2 $mL.kg^{-1}.min^{-1}$ in the final minute of each workload. Steady state was achieved by all of the participants within the set time. Rowland (1996) suggested that the source of the "aerobic drift" observed during prolonged submaximal exercise might be related to temperature regulation, intravascular volume, or changes in muscle fibre recruitment. In agreement with Rowland (1996), Robergs and Roberts (1997) contended that an increase in muscle temperature and circulating catecholamines were identified as sources of aerobic drift during prolonged submaximal exercise.

Studies examining aerobic drift in children (Macek *et al.*, 1976; Asano & Hirakoba, 1984; Rowland & Rimany, 1995) have imposed constant workloads that have represented approximately 60% of the participants' peak oxygen uptake for 40 minutes (Rowland & Rimany, 1995) and 60 minutes (Macek *et al.*, 1976; Asano & Hirakoba, 1984). Rowland and Rimany (1995) reported an aerobic drift of 8.6% between minutes 10 and 40 of constant load submaximal exercise in 11 premenarcheal girls who cycled on a cycle ergometer. Aerobic drift between 10 and 60 minutes of constant load submaximal exercise was observed to be 11% in 11 boys age between 10 and 12 years (Asano & Hirakoba, 1984) and 10.5% in 10 boys aged between 11.6 and 14 years (Macek *et al.*, 1976). The consistently higher oxygen uptake observed during CISP in this study may not be indicative of aerobic drift because the percentage differences in oxygen uptake are much smaller than those observed in the other pediatric studies cited (Macek *et al.*, 1976; Asano & Hirakoba, 1984; Rowland & Rimany, 1995). One possible explanation for the

observed differences may be related to the exercise time (3-minute steady states in the present study) which was shorter than that of the other studies. The shorter exercise time may also have resulted in no differences in the oxygen uptake when DSP and CISP were compared.

5.5.1.1 Differences in Heart Rate

5.5.1.1.1 The Impact of Exercise On Body Heat

Exercise is responsible for producing additional metabolic heat in the body. The body has a variety of mechanisms for dealing with this metabolic heat. One of these mechanisms includes dilating the blood vessels of the skin to augment the dissipation of internal metabolic heat by increasing the blood flow to the skin. This response results in some adjustments to the cardiovascular system in order to compensate for the blood shunting to the skin. In adults, this causes a diminished central blood volume and a consequently a reduced cardiac filling. As a result of the reduced cardiac filling, stroke volume is compromised. Cardiac output is the result of stroke volume and heart rate; therefore when stroke volume is reduced, as it is in this case, the response is to increase heart rate (Rowland, 1996).

The mechanisms of thermal regulation in children are different to those of adults (Rowland, 1996). The response of children to the heat produced during exercise, which is a reflection of increased skeletal muscle metabolism, is either greater, the same, or less when children are compared with adults. The thermal response of children depends on the mode of exercise, its intensity, and the way that the exercise is expressed in relation to body size (Rowland, 1996). When children undertake submaximal exercise on a treadmill (weight-bearing), the heat produced is greater in younger children when compared with their older peers (MacDougall *et al.*, 1983). There are a variety of mechanisms that children employ to maintain thermal homeostasis; one of these mechanisms includes a cardiovascular response (Rowland, 1996). The physiological responses to 50 minutes of treadmill walking in 5 pre-pubertal girls when compared with adult females was examined by (Drinkwater *et al.*, 1977). The girls exercised at 30% of their peak oxygen uptake at three different ambient temperatures and relative humidity.. The girls exhibited a higher mean heart rate and lower stroke volume when compared with the adults. Heart rates over 90% of those recorded at maximal exercise were observed in four of the five girls. The result of the increase in heart rate led to four of the five girls terminating the exercise bout before the planned exercise duration was completed. Drinkwater and colleagues (1977) observed only a small increase in core temperature during the exercise bouts and consequently considered the exercise limitation to be cardiovascular in nature. In the present study, attempts were made to

reduce the thermal load on the participants by having a fan directed at the participants at all times during the submaximal exercise tests.

In the present study, the higher heart rates (Figure 5.3) were observed when CISP was compared with DSP. This observation may be a result of the cardiovascular response to exercise reported by Drinkwater et al., (1977) and Rowland (1996). The heart rates observed during the final two workloads of the CISP (8.0 and 8.5 $km.hr^{-1}$) would have been influenced not only by the weight-bearing nature of inclined treadmill running but also by an incremental increase in exercise intensity. Rowland (1996) identified both of these parameters as a source of greater metabolic heat in young children.

5.5.1.2 Differences in the RER

The observed difference in the RER when DSP and CISP were compared was evident in the final workload of 8.5 $km.hr^{-1}$ (Figure 5.2). The source of the higher RER may be related to a possible increase in body temperature and, perhaps, more directly to a possible increase in muscle temperature. The impact of an increase in muscle temperature during exercise still remains controversial (Koga *et al.*, 1997). Some suggested effects have been an increase in the oxygen uptake (QO_2) of isolated mitochondria by a Q_{10} effect which may decrease the phosphorylation potential of the muscle (Brooks *et al.*, 1971; Willis & Jackman, 1994). Koga et al. (1997) did not find a difference in the RER at the end of 6 minutes of moderate (below Anaerobic Threshold (AT)) and heavy exercise (above AT) in 7 adult males. When the results of the present study were compared with those of Koga et al. (1997), the exercise time for the CISP was double that reported by Koga et al. (1997) and ranged in intensity from 48 - 88% across all of the participants. The exercise intensities investigated by Koga et al., (1997) were duplicated within the CISP for 90% of the participants in this study. The incremental nature of the CISP and the possible cumulative thermal load may have had an influence on the mitochondrial respiration. This, in turn, may have decreased the phosphorylative potential of the mitochondria and driven the increase in RER that was observed in the final workload.

5.5.1.3 Differences in the Ventilation

Differences were observed in ventilation (V_E $L.min^{-1}$) between DSP and CISP (Figure 5.4). The higher V_E observed during CISP may be related to the higher oxygen uptake also observed in the final workload of the CISP. The increase in V_E may be related to the increased oxygen cost of ventilation (Dick and Cavanagh, 1987).

5.5.1.4 The repeatability and variability of oxygen uptake during CISP

The repeatability of oxygen uptake found in the present study compares well with the results of Rogers et al. (1994), Frost et al. (1995), and Unnithan et al. (1995). Rogers et al. (1994) reported a repeatability of 0.84 for 5 mph (≈ 8 $km.hr^{-1}$) and 0.86 for 6 mph (≈ 10 $km.hr^{-1}$). The repeatability reported by Rogers et al. (1994) is similar to that observed in the present study (Table 5.4).

5.5.1.4.1 Mean within-participant variability in oxygen uptake

The mean within-participant variability (individual variability) of oxygen uptake in the present study was slightly higher than that reported by Rogers et al. (1994). The within-participant variability for 5 mph was 4.9% and 4.5% for 6 mph (Rogers *et al.*, 1994). Unnithan (1993) observed a within-participant variability of 4.2%, 5.6% and 5.9% for treadmill speeds of 7.2, 8.0 and 8.8 $km.hr^{-1}$. The within-participant variability reported in the present study is higher for the first submaximal workload, but comparable to those reported for the faster treadmill speeds (Unnithan, 1993). Unnithan (1993) reported increasing between-participant variability with an increase in exercise intensity. These findings are in direct contrast to the findings of the present study of decreasing between-participant variability with increasing exercise intensity.

5.5.1.4.2 Range of within-participant variability in oxygen uptake

Large ranges in within-participant variability for oxygen uptake were reported in the studies conducted by Unnithan (1993) and Rogers et al. (1994) reported ranges of within-participant variability of between 0.3 - 11.5% for the 5 mph and 0.3 - 11.9% for 6 mph. Unnithan (1993) reported a within-participant variability of between 0.3 and 17% at 8.8 $km.hr^{-1}$. A large range of within-participant variability was found for the lowest submaximal intensity in the present study when compared with the findings of Rogers et al. (1994). Smaller ranges in within-participant variability were found at the highest submaximal intensity in the present study when compared with those reported by Unnithan (1993). Frost et al. (1995) also observed several different response patterns in the participant's involved in their study but did not report the ranges.

5.5.1.4.3 Possible sources of large range of between-participant variability in oxygen uptake

Factors such as training status, diet, running mechanics, circadian variation, ambient temperature, footwear, and length of time for treadmill habituation have been suggested as sources of large ranges of within-participant variability of oxygen uptake (Williams & Armstrong, 1991). In the present study, factors such as circadian variation, ambient temperature, and footwear were

controlled. The participants attended the laboratory for testing at the same time each day and the ambient temperature was between 20 and 22^{0}C. Each participant wore the same footwear in both tests. Other factors such as training status, diet, and running mechanics were not controlled for the present study. All of the participants were given the same time to acclimatise to the treadmill according to the protocol described in the methodology section of this thesis; however, two participants in the present study had previous treadmill running experience prior to the conduct of the study. Between participant variability may be related to maturity differences in children (Freedson *et al.*, 1981; Sady *et al.*, 1989). Sady and Katch (1981) and Sady et al. (1989) observed that the highest between-participant variability occurred in the less mature individual. Although the participants in the present study were all pre-pubertal, it is possible that some participants were more mature (closer to puberty) than others. This may have affected the range of between-participant variability in oxygen uptake.

5.5.1.5 The repeatability and variability of heart rate during CISP

No differences in the heart rate recorded during CISP during two testing sessions over three days are in agreement with other studies examining heart rate during submaximal exercise across two separate testing sessions (Rogers *et al.*, 1994; Frost *et al.*, 1995; Unnithan *et al.*, 1995).

The heart rate of the participants in the present study was higher in the second testing session when compared with the first. The largest difference in heart rate occurred at the first submaximal intensity and then decreased throughout the testing. This may have been because the participants were anxious about the test, or due to a biological variation in heart rate. These findings are in contrast to the findings of Frost et al. (1995) who found a decrease in heart rate during the second testing session when compared with that of the first testing session.

5.5.1.5.1 Mean and range of within-participant variability in heart rate

The mean within-participant variability of heart rate in the present study was similar to that observed by Rogers et al. (1994). The range was greater at the first submaximal intensity in the present study when compared with that of Rogers et al. (1994). In the second and third submaximal intensities, the range decreased. This observation is similar to that of Rogers et al. (1994) although, the range is smaller in the present study when compared with that of Rogers et al. (1994).

5.5.2 *Using DSP and CISP for the determination of the supramaximal workload.*

No differences were found in the linear regression parameters used to predict the supramaximal workloads. Green and Dawson (1996a) also examined the use of DSP and CISP on the prediction of supramaximal exercise workloads using a cycle ergometer. The results of this study showed no differences in the mean regression equations used to predict the supramaximal workload when the DSP and CISP were compared (Green and Dawson, 1996a). The results of this study are in agreement with those found in the study of Green and Dawson (1996a).

5.5.3 *Comparison of AOD with different submaximal protocols*

There were no differences in AOD of the participants when either of the submaximal exercise protocols was used to determine the workload for the supramaximal test. AOD ($\sum$L and mL.kg^{-1}) was greater in the DST when compared with the CST. The supramaximal test time was also greater in the DST when compared with the CST. The longer test time in the DST may be related to the slower treadmill speed when compared with the treadmill speed of the CST. The participants may have been able to run longer during the DST because the exercise intensity was not as great.

When AOD are compared with those of the pre-pubertal participants in the first study (Study One) in this thesis, the AOD values ($\sum$mL.kg^{-1}) from the DST are larger when compared with Study One. The AOD values ($\sum$mL.kg^{-1}) are smaller when the CST is compared with study one. AOD reported in the present study are within the range of values reported by Naughton and Carlson (1998) for pre-pubertal males using the treadmill as the mode of exercise.

This study showed that there was no difference in the oxygen uptake when DSP and CISP were compared. There was also no difference in the linear regression equations constructed from the relationship between oxygen uptake and exercise intensity during submaximal exercise and no difference in AOD when the two submaximal protocols were compared. The second part of the study showed that oxygen uptake during CISP was reliable and suggests that the use of a CISP may be useful in determining the submaximal relationship and may be less time-consuming.

Chapter 6

STUDY THREE

A COMPARISON OF ACCUMULATED OXYGEN DEFICIT WITH RUNNING AND CYCLING EXERCISE IN PRE-PUBERTAL MALES

A COMPARISON OF ACCUMULATED OXYGEN DEFICIT WITH RUNNING AND CYCLING EXERCISE IN PRE-PUBERTAL MALES

Abstract

Traditionally, accumulated oxygen deficit has been measured using either running or cycling exercise. Differences in the magnitude of accumulated oxygen deficit (AOD) have been observed with different modes of exercise. Running exercise has been shown to produce a larger AOD than cycling exercise. It has been postulated that running exercise yields a larger AOD due to the larger active muscle mass associated with running exercise. To date, no studies have compared the AOD with running and cycling exercise using the same sample of participants. Consequently, the purpose of this study was to compare the difference in AOD with running and cycling exercise. Ten pre-pubertal males (mean age 11.9 ± 0.49) who volunteered to participate in the study attended the laboratory on four occasions. During the study, the participants completed randomised peak oxygen uptake tests on the motorised treadmill and cycle ergometer. Continuous incremental submaximal exercise tests were also conducted on both the treadmill and cycle ergometer to determine the relationship between submaximal oxygen uptake and exercise intensity. Individual linear regression equations were constructed to determine the relationship between submaximal oxygen uptake and exercise intensity and to predict a supramaximal exercise intensity that would equate to 120% peak oxygen uptake on both the treadmill and cycle ergometer. AOD was calculated for both exercise modes using the difference between the predicted oxygen demand and the actual oxygen uptake for the duration of the supramaximal test. In order to compare two different measures of anaerobic performance, each participant completed a Wingate Anaerobic Test (WanT). The parameters of the WAnT were correlated with AOD on the cycle ergometer to determine whether there was any relationship between the two tests. A one-way analysis of variance (ANOVA) was used to determine whether there was a difference in AOD when running and cycling exercise were compared. Pearson correlation coefficients were used to determine whether there was any relationship between the WAnT and AOD. ANOVA revealed that AOD was larger when running (54.68 ± 3.67 $mL.kg^{-1}$) and cycling exercise (33.96 ± 4.42 $mL.kg^{-1}$) were compared ($p < 0.05$). AOD may be greater due to the larger active muscle mass engaged during running exercise when compared with cycling exercise. There was also a strong correlation between peak power (watts) and AOD ($\sum L$) ($p = 0.002$). These findings suggest that the AOD may be an acceptable measure of anaerobic performance comparable with the universally accepted WAnT.

6.1 Introduction

Traditionally, accumulated oxygen deficit (AOD) has been measured using either running or cycling as the modes of exercise (Medbø et al. 1988; Medbø and Tabata, 1989; Ramsbottom et al. 1994; Gastin and Lawson 1994b; Green and Dawson, 1995; Hill, 1996; Pizza et al. 1996). Other modes of exercise such as swimming and kayaking have also been used to determine the AOD (Bazdukas *et al.*, 1991; Terrados *et al.*, 1991). Gastin (1994) observed that AOD appeared to be related to the size of the active muscle mass. Gastin (1994) compared studies using a variety of exercise modes and suggested that AOD was larger with running when compared with cycling exercise. Smaller AOD's have been observed with kayaking and swimming exercise (Bazdukas et al. 1991; Terrados et al. 1991; Troup et al. 1991). Gastin (1994) suggested that AOD was even smaller with kayaking and swimming , when compared with running and cycling exercise, as a result of the smaller active muscle mass involved in the exercise. Only one author to date has compared AOD in two different modes of exercise using the same participants (Bangsbo *et al.*, 1993). Bangsbo et al. (1993) compared AOD in five rowers who ran on the treadmill and rowed on a rowing ergometer. The authors found that AOD was higher with rowing when compared with running and attributed the larger AOD with rowing exercise to the greater muscle mass involved in the exercise (Bangsbo et al., 1993).

The results of Studies Two and Three of this thesis have indicated that a continuous incremental submaximal protocol may be useful in determining the relationship between submaximal oxygen uptake and exercise intensity. The continuous incremental submaximal protocol is more time efficient than the traditional method of determining the relationship between submaximal oxygen uptake and exercise intensity using discrete submaximal exercise tests (Medbø, et al., 1988). Consequently, an examination of the possible difference in accumulated oxygen deficit with different modes of exercise in the same sample population was deemed to be achievable. Another consideration of this study was to investigate whether there was a relationship between a universally accepted measure of anaerobic performance, the Wingate Anaerobic Test (WAnT), and AOD. To date, no comparisons between these two measures of anaerobic performance have been made in pediatric populations.

6.2 Purpose

The purpose of this study was to compare accumulated oxygen deficit in a sample of pre-pubertal males using treadmill running and cycling as the modes of exercise. In order to investigate whether there was a relationship between AOD and another measure of anaerobic performance, the association of the WanT and AOD were also undertaken.

6.3 Methodology

The methodology employed during this study will be presented in 6 major sections: the participants; the continuous incremental submaximal exercise test protocols; peak oxygen uptake measures; supramaximal exercise tests; the determination of sexual maturation; and, the statistical examination of the results.

6.3.1 The participants

Ten pre-pubertal males (mean age 11.9 ± 0.49) volunteered to participate in this study. All of the participants who volunteered for this study were trained runners. Prior to the commencement of this study, all participants came to the laboratory for a familiarisation session. The conduct of the familiarisation session has been described previously (Chapter 3). The participants attended the laboratory for testing on four occasions.

6.3.2 Peak oxygen uptake measures

Each of the participants completed peak oxygen uptake tests on both the cycle ergometer and the motorised treadmill. The peak oxygen uptake test conducted on the treadmill was conducted in the same way as previously described (Chapter 3). The peak oxygen uptake test for cycling was conducted on a CYBEX™ (met, 100) isokinetic cycle ergometer. The commencing workload was 25 watts. The workload increased by 25 watts every minute until volitional exhaustion. The pedalling cadence was set at 90 revolutions per minute (r.p.m.). The CYBEX™ (met 100) isokinetic cycle ergometer had a display screen which allowed the exercising participant to see their workload, pedal cadence and elapsed time for the duration of the test. Volitional exhaustion was determined according to the criteria set by Zwiren (1989).

6.3.3 Continuous incremental submaximal exercise protocols

Continuous incremental submaximal exercise tests were conducted on a motorised treadmill and cycle ergometer. The continuous incremental submaximal exercise tests were completed in a random order. Each participant completed at least four workload increments during the continuous incremental test on either the treadmill or the cycle ergometer. Each submaximal workload was between three and four minutes in duration. The participants were required to reach steady state oxygen uptake (according to the definition in Chapter 3) (± 2mL.kg^{-1}.min^{-1}) at each submaximal intensity before proceeding to the next workload. At the completion of the continuous incremental submaximal exercise test, the participants rested for at least 30 minutes before commencing the peak oxygen uptake test on the same piece of equipment.

6.3.4 Supramaximal exercise tests

Supramaximal exercise tests were conducted on the treadmill and cycle ergometer at workloads that represented 120% of peak oxygen uptake determined during the respective peak oxygen uptake test. Supramaximal workloads were calculated using individual linear regression equations that described the relationship between submaximal oxygen uptake and exercise intensity in the method described previously (Chapter 3, Appendix 3). The supramaximal tests were conducted in the method described in the methods section (Chapter Three). The supramaximal test conducted on the cycle ergometer was terminated when the participant was no longer able to maintain a pedal cadence above 60 r.p.m. The supramaximal test conducted on the treadmill was terminated when the participant was no longer able to maintain the treadmill speed.

6.3.4.1 Wingate Anaerobic Tests

Each participant completed a 30-second all-out cycle test on the isokinetic ergometer (CYBEX™, met 100). During this test, peak and mean power was provided every two-seconds (CYBEX™, Met emulator software, Ball State University, 1991). Support for the use of an isokinetic cycle ergometer for all-out cycle tests with the pediatric population originates from the research of Naughton (1993). Naughton (1993) observed that isokinetic devices such as the CYBEX™ (Met, 100) required the participant to overcome initial inertial forces, but that once this had been achieved, there would be no variation in the angular acceleration once the isokinetic velocity was reached. Consequently, the accelerations and decelerations of the participant, which may be a result of fatigue, may be more stable on an isokinetic cycle ergometer. Sargeant (1989) supported the use of accommodating resistance, such as that offered by isokinetic devices, for the pediatric population because he believed that they might enhance the selection of optimal testing conditions.

6.3.5 Determination of sexual maturation

During the testing period, the participant's parents were asked to identify their child's level of sexual maturity according to the Tanner Scale (Tanner, 1962). In the first instance parents were informed of the use of the Tanner Scale as an indicator of sexual maturity in the Parent's Letter (Appendix 1.5). Once the study commenced, the parents were sent a letter alerting them to the fact that the Tanner Scale pictorial representations would be forwarded to them in the mail in the following week. They were also encouraged to contact the investigator if there were any queries. The Tanner Scale pictorials were then forwarded to the parents with instructions on how to

complete the task (Appendix 1.5). Once the replies were received back from the parents, the Tanner Scale stage was recorded and the letters were shredded.

6.3.6 Statistical analysis of the results

The descriptive data was expressed as means ± standard errors (Mean ± S.E.M.). One-way analysis of variance (ANOVA) was used to determine the difference in accumulated oxygen deficit when running and cycling exercise were compared. Pearson correlation coefficients were used to determine whether there were any relationships between the parameters from the Wingate Anaerobic Test and accumulated oxygen deficit. The alpha level of 0.05 was adopted for all testing.

6.4 Results

The focus of this study was to compare accumulated oxygen deficit with running and cycling exercise in pre-pubertal males. The results of this study will be presented in three major sections: participants' profiles; a comparison of accumulated oxygen deficit measures with running and cycling exercise; and, Wingate Anaerobic Test results.

6.4.1 Participant profiles

6.4.1.1 Descriptive characteristics

The descriptive characteristics of the sample group are presented in Table 6.1. The participants were of average mass (50^{th} percentile) and average height (50^{th} percentile) when compared with a sample population of caucasian males of the same age (NHANES, 1992).

Table 6.1 Descriptive characteristics

Age (yr)	Mass (kg)	Height (cm)
11.94	39.23	150
± 0.49	± 2.26	± 2

Mean ± S.E.M.

6.4.1.2 Maximal effort profiles

The maximal effort profiles of the participants who completed this study are presented in Table 6.2. (Appendix 6.1) Two maximal effort profiles were conducted in this study, one on a motorised treadmill, the other on a cycle ergometer. Relative peak oxygen uptake ($mL.kg^{-1}.min^{-1}$) was significantly greater when the treadmill peak oxygen uptake test was compared with the cycle ergometer peak oxygen uptake test ($p = 0.001$). The treadmill peak oxygen uptake test elicited a significantly greater maximal heart rate when compared with the cycle ergometer peak oxygen uptake test ($p = 0.027$). In contrast, the RER values for the cycle ergometer peak oxygen uptake test were significantly greater than those of the treadmill peak oxygen uptake test ($p = 0.008$). In percentage terms, the absolute peak oxygen uptake ($L.min^{-1}$) was 10.7% higher during the treadmill peak oxygen uptake test when compared with that of the cycle ergometer peak oxygen uptake test. The relative peak oxygen uptake ($mL.kg^{-1}.min^{-1}$) during the treadmill peak oxygen uptake test was 26.9% greater than the cycle ergometer peak oxygen uptake test. The treadmill peak oxygen uptake test also elicited a 4.5% greater maximal heart rate when compared

with the cycle ergometer peak oxygen uptake test. This trend was reversed when the RER values were compared in percentage terms, the cycle ergometer peak oxygen uptake test having elicited a 5.3% greater RER than the treadmill peak oxygen uptake test.

Table 6.2 Maximal effort profiles

	Treadmill	Cycle Ergometer
Peak oxygen uptake ($L.min^{-1}$)	2.66 ± 0.19	2.21 ± 0.17
(range)	(1.69 - 3.66)	(1.31 - 3.02)
Peak oxygen uptake ($mL.kg^{-1}.min^{-1}$)	67.64 ± 2.03 [a]	56.22 ± 1.98
(range)	(55.15 - 75.46)	(43.57 - 65.27)
Peak heart rate ($b.min^{-1}$)	201 ± 2 [b]	192 ± 3
Peak RER	1.09 ± 0.01	1.15 ± 0.01 [c]

Mean ± S.E.M.

'a' denotes treadmill different from cycle ergometer ($p < 0.05$)

'b' denotes treadmill different from cycle ergometer ($p < 0.05$)

'c' denotes cycle ergometer different from treadmill($p < 0.05$)

All of the participants attained maximal performances by fulfilling at least one of the criteria suggested by Zwiren, (1989). These criteria (Zwiren, 1989) include either a peak heart rate that was 95% of their age-predicted maximum, a respiratory exchange ratio (RER) in excess of 1.05, or a leveling off of oxygen uptake with increasing workload. During the treadmill peak oxygen uptake test, sixty percent of the participants fulfilled the heart rate criteria as well as the RER criteria; ninety percent achieved the RER criteria and none of the participants experienced a plateau in oxygen uptake with increasing workload. In contrast, during the cycle ergometer peak oxygen uptake test, only thirty percent of the participants fulfilled the heart rate criteria as well as the RER criteria, one hundred percent achieved the RER criteria and none of the participants experienced a plateau in oxygen uptake with increasing workload. The observation that none of the participants experienced a leveling-off of oxygen uptake with increasing workload, supports the findings of Rowland (1996) that the majority of children in younger populations may not exhibit a leveling-off of oxygen uptake with increasing workload; consequently, it was not used as a criteria for achieving peak oxygen uptake.

6.4.2 *Submaximal exercise and the prediction of the supramaximal workload*

The following section presents results of mean submaximal exercise intensities and mean linear regression equations.

6.4.2.1 Submaximal exercise intensity comparisons

Table 6.3 presents the mean values for the lowest exercise intensity and the highest exercise intensity expressed as a percentage of the peak oxygen uptake for submaximal tests conducted on the treadmill and cycle ergometer. Submaximal steady state exercise was conducted on a motorised treadmill and a cycle ergometer as described in the methodology section of this chapter. Each participant completed between three and four submaximal continuous incremental steady exercise bouts on the treadmill and the cycle ergometer. Participants completed continuous incremental submaximal exercise bouts on the treadmill at speeds ranging from 6 – 12 $km.hr^{-1}$. Participants also completed continuous incremental submaximal exercise bouts on the cycle ergometer at workloads ranging from 25 - 175 watts.

Table 6.3 Mean and range peak submaximal oxygen uptake (%) on treadmill and cycle ergometer

	Treadmill	**Cycle Ergometer**
Mean lowest % oxygen uptake	54.43 ± 1.98	47.43 ± 3.52
(range)	(45.6 - 64.0)	(36.0 - 71.0)
Mean highest % oxygen uptake	81.06 ± 1.16	82.88 ± 3.37
(range)	(77.4 - 88.6)	(69.0 - 96.9)

Mean ± S.E.M.

There was no difference between the mean lowest percent oxygen uptake when the treadmill and cycle ergometer were compared ($p = 0.101$) (Appendix 6.2). The same trend was observed when the mean highest percent oxygen uptake values for submaximal treadmill exercise were compared with submaximal cycle ergometer exercise ($p = 0.617$) (Appendix 6.2). A larger range of submaximal exercise intensities was observed when the cycle ergometer was compared with the treadmill. The range of mean percent lowest oxygen uptake was 18.4% for treadmill exercise and 35.0% for the cycle ergometer. The same trend was observed when the range of mean percent highest oxygen uptake between treadmill and cycle ergometer. The range of treadmill values was 11.2% and the range for the cycle ergometer was 27.9%. Medbø and Tabata (1989) proposed that steady state submaximal exercise be conducted at intensities that represented 30 - 100% of

the participant's peak oxygen uptake. The mean lowest and highest submaximal exercise intensities fell within the range suggested by Medbø and Tabata (1989).

6.4.2.2 Mean linear regression equations constructed to predict supramaximal workload

The mean linear regression equations for treadmill and cycle ergometer submaximal exercise bouts are presented in Table 6.4. Subsequent to the conduct of the submaximal steady state tests individual regression analyses were performed. The regression analyses were of linear form [y = a + b(x)] and described the relationship between submaximal oxygen uptake and exercise intensity. In order to predict the supramaximal workload the linear regression equation was rearranged so that [y = a + b(x)] became (x) = (y - a)/b where y represents oxygen uptake and (x) represents the exercise intensity (see Appendix 3 for an example calculation). The resultant y intercept (a) and slope of the regression line (b) were used to predict the supramaximal workload that represented 120% of the participants' maximal oxygen uptake.

Table 6.4 Mean linear regression equations

	Treadmill	**Cycle Ergometer**
Y Intercept	0.14 ± 0.00	0.30 ± 0.01
Slope	5.24 ± 0.82	103.5 ± 0.33
Pearson Correlation (r)	0.98	0.98
Accountable variance (R^2)	96%	96%
R^2 x 100		

Mean ± S.E.M. Note that regression equation for treadmill is in $mL.kg^{-1}.min^{-1}$ and for cycle in $l.min^{-1}$

The accountable variance represents the variance in the confidence in the linearity of the regression line. These regression lines express the relationship between continuous incremental submaximal oxygen uptake and exercise intensity; therefore the accountable variance represents the variance in continuous incremental submaximal oxygen uptake that can be accounted for by the submaximal exercise intensity. Ninety-six percent of the variance in submaximal oxygen uptake is accounted for by the exercise intensity for both the treadmill and cycle ergometer.

The linear regression equations were then used to predict the exercise intensity that represented 120% of the participant's peak oxygen uptake (supramaximal exercise). As treadmill running and bicycling were the modes of exercise used for this study, treadmill speeds and resistances for the cycle ergometer were predicted from the linear regression equations. For an example of this

calculation, see Appendix 3. The mean treadmill speed required to elicit 120% of peak oxygen uptake for the participant's was 15.4 ± 0.88 km.hr^{-1} (range 9.03 - 19.8 km.hr^{-1}.). The mean resistance required to elicit 120% of peak oxygen uptake for the participant's was 227.03 ± 18.57 watts (range 155.0 - 319.0 watts).

6.4.3 Measures of accumulated oxygen deficit

The supramaximal test parameters and AOD calculated from the supramaximal tests are presented in three sections: the supramaximal tests; relative aerobic and anaerobic contributions to the supramaximal tests; and, measures of accumulated oxygen deficit.

6.4.3.1 The supramaximal tests

Table 6.5 presents the mean supramaximal test parameters: supramaximal test time, supramaximal heart rate and supramaximal RER for the supramaximal tests conducted on the treadmill and the cycle ergometer. The two supramaximal tests were conducted in random order.

Table 6.5 Supramaximal test parameters

	Treadmill	**Cycle Ergometer**
Supramaximal test time (sec)	126.54 ± 19.93[a]	61.79 ± 6.94
Supramaximal heart rate (b.min-1)	190 ± 3	182 ± 4
Supramaximal RER	1.02 ± 0.01	0.95 ± 0.03

Mean ± S.E.M.

'a' denotes treadmill different from cycle ergometer ($p < 0.05$).

The supramaximal test conducted on the treadmill was 50.2% longer than the supramaximal test conducted on the cycle ergometer. The supramaximal test conducted on the treadmill also elicited a higher supramaximal heart rate (4.3%) and a higher RER (6.9%) when compared with the supramaximal test conducted on the cycle ergometer.

6.4.3.2 The relative aerobic and anaerobic contributions (%) to the treadmill and cycle ergometer supramaximal test

The relative aerobic and anaerobic contributions (%) to the supramaximal exercise tests on the treadmill and cycle ergometer are presented in Figure 6.1. (Appendix 6.4) The relative aerobic and anaerobic contributions to the supramaximal exercise tests on the treadmill and cycle ergometer were calculated by using accumulated oxygen demand. Estimations of the aerobic and

anaerobic contributions to both of the exercise tests were made by dividing the actual O_2 consumed during the supramaximal exercise test by the predicted accumulated oxygen demand. This gave an estimation of the relative aerobic contribution to the supramaximal exercise test.

The estimated anaerobic contribution to the supramaximal exercise test on the treadmill was 35.5%. In comparison, the estimated anaerobic contribution to the supramaximal exercise test conducted on the cycle ergometer was 45.1%. These differences represent an estimated 21.3% greater contribution from the anaerobic system during the cycle ergometer supramaximal test when compared with the treadmill supramaximal test.

6.4.3.3 Accumulated oxygen deficit measures

The anaerobic performance characteristics measured by AOD are presented in Table 6.6 and Figures 6.2 and 6.3. (Appendix 6.5) There were differences ($p = 0.013$) in the AOD (ΣL) when running on the treadmill and cycling on the cycle ergometer were compared. The absolute (ΣL) AOD was 47.2% greater when of the participants ran compared with when they cycled. When the AOD ($\Sigma mL.kg^{-1}$) values were compared there was a difference ($p = 0.002$) when running and cycling. This difference represented a 37.9% larger AOD ($\Sigma mL.kg^{-1}$) when running and cycling were compared.

Table 6.6 AOD for running and cycling

	Running	Cycling
AOD (ΣL)	2.25 ± 0.32^a	1.19 ± 0.20
AOD ($\Sigma mL.kg^{-1}$)	54.68 ± 3.67^b	33.96 ± 4.42

Mean ± S.E.M.

'a' denotes running different from cycling ($p < 0.05$).

'b' denotes running different from cycling ($p < 0.05$).

6.4.4 Wingate Anaerobic Test (WAnT)

All participants completed a WAnT on the isokinetic cycle ergometer. The results of the WAnT are presented in Table 6.7. The range of peak power generated during the WAnT was 218 - 511 watts. This range represents a 57.4% difference in the highest peak power and the lowest peak power. The range of mean power generated during the WanT was 188.5 - 419 watts. The range of mean power represents a 55.1% difference in the highest and lowest mean power. The percentage

difference in the range of relative peak power (watts/kg^{-1}) was smaller (39.9%) than that of peak and mean power.

Pearson correlation coefficients were computed in order to investigate whether there was a relationship between the WAnT and AOD measures when cycling was the mode of exercise. A strong correlation (0.846) was observed between WAnT peak power (watts) and AOD ($\sum$L) for cycling ($p = 0.002$). A medium correlation (0.739) was observed between relative peak power (watts/kg^{-1}) and AOD ($\sum$L) ($p = 0.015$)(Appendix 6.6).

Table 6.7 Wingate Anaerobic test parameters

Peak Power (watts)	Mean Power (watts)	Relative Peak Power (watts/kg^{-1})	Maximal Heart Rate (b.min^{-1})
311.00	269.55	7.77	178
± 30.95	± 22.28	± 0.33	± 2

Mean ± S.E.M.

Figure 6.1 Relative aerobic and anaerobic contributions to the supramaximal test

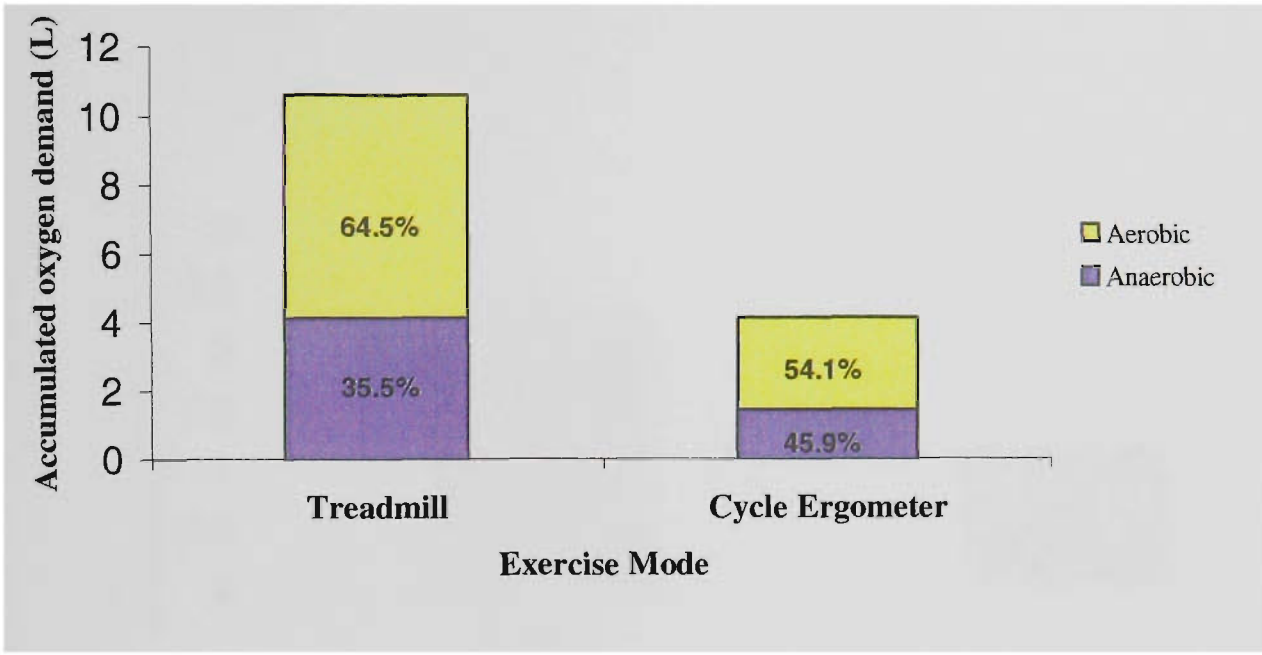
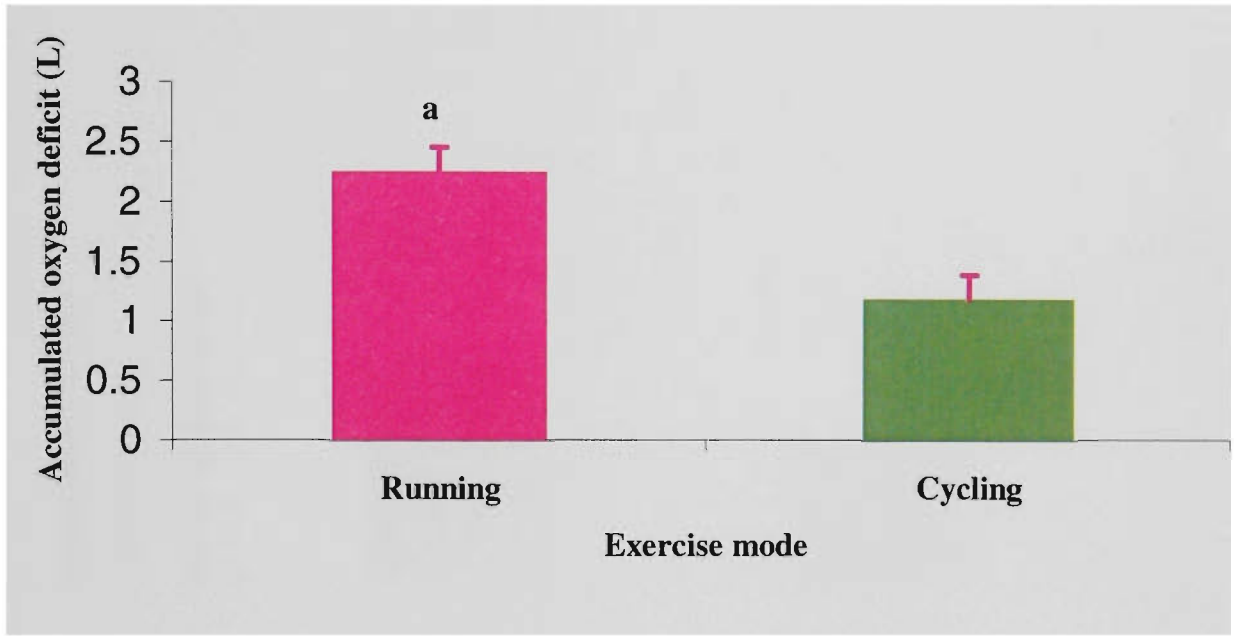


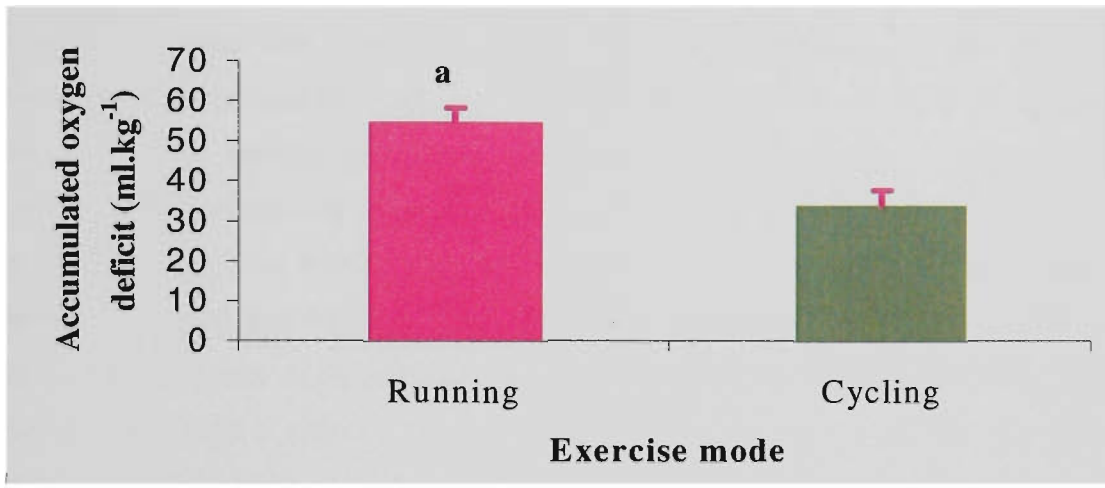
Figure 6.2 AOD (ΣL) comparisons of running and cycling AODs



Mean ± S.E.M.

'a' denotes running different from cycling

Figure 6.3 AOD (Σml. Kg^{-1}) comparisons between running and cycling



Mean ± S.E.M.

'a' denotes running different from cycling

6.5 Discussion

This study compared AOD with running and cycling exercise in pre-pubertal males. This study found significant differences in the absolute (ΣL) and relative (ΣmL.kg^{-1}) AOD when running and cycling exercise were compared.

6.5.1 Possible reasons for the differences in AOD with running and cycling exercise.

To date, there have been no published studies investigating the differences in AOD after running or cycling exercise with either adult or pediatric populations (Gastin, 1994). AOD appears, however, to be related to the size of the exercising muscle mass (Medbø et al. 1988; Medbø and Tabata, 1989; Medbø and Burgers, 1990; Bangsbo, et al. 1993; Weyand et al. 1993; Withers et al. 1993; Pizza et al. 1996). Medbø and Tabata (1989) reported a 30% larger AOD in adults after 2 minutes of exercise when they compared inclined treadmill running with cycling. These results (1989) were drawn from separate studies. Medbø and Tabata (1989) attributed the difference in AOD with running and cycling exercise to different use of the leg muscles. A larger AOD when running and cycling exercise was compared (Medbø and Tabata, 1989) was also observed in the present study. The difference in AOD is greater (37.9%) in the present study when compared with that observed by Medbø and Tabata (1989). The larger difference in the present study may be explained by the difference in the supramaximal test time. Medbø and Tabata (1989) compared differences in AOD after 2 minutes of running and cycling exercise. In the present study, the participants completed a mean of 2.06 minutes of running and 1.01 minutes of cycling exercise. Supramaximal test time has been seen to affect AOD. Medbø and Tabata (1989) observed that there was a significant increase in AOD with increasing exercise time. A supramaximal test time that is between 2 and 3 minutes has been proposed as ideal for determining AOD (Medbø and Tabata, 1989). In the present study, the mean supramaximal test time on the cycle ergometer was just over 1 minute which is a minute short of the 2 - 3 minutes proposed by Medbø and Tabata (1989) as ideal. As a result of the shorter supramaximal test time on the cycle ergometer, AOD for cycling may be underestimated in the present study. The supramaximal test on the cycle ergometer was concluded when the participant was no longer able to maintain a pedal cadence above 60 revolutions per minute. The supramaximal heart rate and RER at the conclusion of the supramaximal test conducted on the cycle ergometer, were lower than those observed at the conclusion of the supramaximal test conducted on the treadmill. All of these factors suggest that the supramaximal test conducted on the cycle ergometer may have concluded prior to exhaustion. The participants in the present study reported that they experienced significant local fatigue in the leg muscles during the supramaximal test conducted on the cycle ergometer that was not apparent

to the same extent during the treadmill supramaximal test. All of the participants in the present study were trained runners, which may have resulted in a greater ability to tolerate the fatigue they experienced during the supramaximal treadmill test when compared with the cycle ergometer test.

Other studies that support the effect of the mass of the exercising muscle have observed smaller AODs from running exercise in females when compared with males (Medbø and Burgers, 1990). Larger AODs from running exercise have also been observed with higher treadmill gradients (Medbø and Burgers, 1990; Olesen, 1992). Medbø and Burgers (1990) suggested that larger AODs observed with a higher treadmill inclination (10% vs. 5%) might be due to a larger muscle mass being activated at the higher inclination.

Weyand et al. (1993) observed a larger AOD with two-legged cycle exercise when compared with that of one-legged cycle exercise in adult males and females. The larger AOD observed with two-legged cycle exercise was attributed to a greater active muscle mass with two-legged cycling when compared with one-legged cycling. Weyand et al. (1993) supported these findings with a strong correlation between AOD and the estimated active muscle mass ($r = 0.94$).

6.5.2 Measures of anaerobic performance in children: Comparisons with other pediatric studies

The present study investigated the anaerobic performances of pre-pubertal males using AOD and the Wingate Anaerobic test. When comparing the results of the present study with those of other studies, it is important to note that there are differences in study methodologies.

6.5.2.1 Accumulated oxygen deficit

AOD for running and cycling exercise from the present study compare well with those investigating running or cycling exercise in the pediatric population (Carlson & Naughton, 1998). In the present study, AOD calculated from running is greater than that observed in pre-pubertal males in Studies One and Two of this thesis. This may be because the participants in the present study were competitive runners.

6.5.2.2 The Wingate Anaerobic Test

The results of the present study used an isokinetic cycle ergometer. The majority of studies investigating the anaerobic power of children have used a constant load, which is calculated according to the body weight of the participant (Inbar & Bar-Or, 1986; Carlson & Naughton, 1994; Hebestreit *et al.*, 1996). The peak power recorded in the present study (mean 311 watts)

was higher than that reported by Inbar and Bar-Or (1983) which reported a mean peak power of 250 watts and Hebestreit et al. (1996) (mean 260 watts). The mean power in the present study (269 watts) is also higher than that observed by Inbar and Bar-Or (1989) (mean 200 watts). Naughton (1993) used an isokinetic cycle ergometer to evaluate the anaerobic performance of pre-pubertal males. The peak and mean power values observed in the present study are very similar to those reported by Naughton (1993). The relative peak power values observed in the present study (mean 7.7 watts/kg) were lower than (mean 8.2 watts/kg) reported by Carlson and Naughton (1993).

6.5.2.3 Comparing AOD with another measure of anaerobic performance: The Wingate Anaerobic Test

The WAnT has been compared with AOD in adults in the past (Scott et al., 1991). Scott et al. (1991) found a strong correlation with AOD (∑mL.kg-1) and WAnT peak power ($W.kg^{-1}$) ($r = 0.70$, $p<0.01$). In contrast to the findings of Scott et al. (1991), this study found a strong association between peak power and AOD (∑L) and no association between WAnT and AOD ($\sum mL.kg^{-1}$).

In conclusion, this study found that AOD is greater when running is compared with cycling exercise. This may be related to the active muscle mass during exercise. Peak power during the Wingate Anaerobic test is also strongly correlated with AOD (ΣL).). A strong relationship between AOD (L) and peak power may make the AOD a comparable measure to the universally accepted Wingate Anaerobic Test.

Chapter 7

SUMMARY AND RECOMMENDATIONS

7.1 Introduction

The following chapter presents a summary of the findings of each of the three studies that comprise this thesis. Recommendations based on the findings of each of the studies have also been made.

7.2 Summary and recommendations from Study One

In Study One, anaerobic performance was measured in three groups of male children using accumulated oxygen deficit. The three groups of male children represented the pre-pubertal, pubertal and post-pubertal stages of development. A sub-sample of each of the groups agreed to venous blood sampling, which enabled the examination of blood borne indicators of anaerobic metabolism. The blood borne indicators of anaerobic metabolism examined were blood lactate and pH.

The results of this study found that the anaerobic performance of the post-pubertal group was larger than that of both the pubertal and post-pubertal groups. This finding suggests that the anaerobic performance of the pre-pubertal, pubertal and post-pubertal males could be quantified by AOD. The blood borne indicators of anaerobic metabolism blood lactate and pH showed activation of anaerobic metabolism and supported the use of AOD as a measure of anaerobic performance. The post-pubertal group exhibited a greater decline in pH concentration when compared with the pre-pubertal group. The increase in the concentration of blood lactate also showed a greater increase when compared with that of both the pre-pubertal and pubertal groups.

Table 7.1 presents a summary of the findings and recommendations of the first study of this thesis. The major focus of Study One was to determine whether the anaerobic performance of a cross-section of pre-pubertal, pubertal and post-pubertal males could be quantified by the AOD. The major finding was that the AOD ($\sum$L) was able to differentiate between the anaerobic performances of the three cross-sectional developmental groups. A recommendation for further research in this area is to investigate the anaerobic performance of male children using AOD in a longitudinal research design.

The second major finding of this study relates to AOD ($\sum$mL.kg^{-1}) not being able to differentiate between the anaerobic performances of the pubertal and post-pubertal groups. In order to investigate this, future research could focus on the impact of training and pubertal status on anaerobic performance quantified using AOD. It is considered a limitation of this study that training status and the lack of information in relation to the activity levels of the subjects was not documented as it appears as though training status may have masked maturity effects in relation

to AOD ($\sum mL.kg^{-1}$). It is suggested that studies investigating the impact of training and the role of pubertal development on anaerobic performance also take particular care with quantifying and tracking training status and break down circum-pubertal development into its constituent Tanner genital phases 2, 3 and 4 to better differentiate the participants involved in the study.

Recommendation three from the first study proposes that a less time-consuming protocol for determining the relationship between submaximal oxygen uptake and exercise intensity be developed. The present methodology requires that the participant attend the laboratory on at least six occasions. The majority of time spent attending the laboratory is taken up with submaximal exercise protocols. The time-consuming nature of the testing potentially restricts participant recruitment, as many parents are not willing to commit to several visits to the laboratory for testing. Less time-consuming protocols may make determining the anaerobic performance of children using AOD more attractive to researchers.

Table 7.1 Summary of the major findings and recommendations from Study One

Summary of major findings	Recommendations
1. AOD ($\sum L$) was able to differentiate between the anaerobic performances of a cross-section of pre-pubertal, pubertal and post-pubertal males.	1. Further research into the developmental aspects of anaerobic performance using a longitudinal design should be conducted.
2. AOD ($\sum mL.kg^{-1}$) was not able to differentiate between the anaerobic performance of pubertal and post-pubertal males.	2. Conduct research to investigate the impact of training and pubertal development on anaerobic performance measured using AOD. These studies should take particular note of tracking training and circum-pubertal status.
3. Submaximal testing to determine the relationship between submaximal oxygen uptake and exercise intensity was time-consuming and required a large number of visits to the laboratory.	3. Future studies should focus on developing a less time-consuming method of determining the relationship between submaximal oxygen uptake and exercise intensity for predicting the supramaximal workload.
4. Blood borne indicators of anaerobic metabolism reflected the anaerobic performances of the pre-pubertal, pubertal and post-pubertal groups.	4. Incorporate an investigation of blood borne indicators of anaerobic metabolism into a longitudinal study of anaerobic performance.

Blood borne indicators of anaerobic metabolism reflected the anaerobic performances of the different developmental groups. Therefore, the final recommendation suggests that future studies continue to incorporate the use of blood borne indicators of anaerobic metabolism as they can further add to the understanding of the impact of anaerobic work on the developing child.

7.3 Summary and recommendations of Study Two

Study Two was examined a method of enhancing and expediting the measurement of AOD. In order to contribute to this aim Study Two investigated the difference in oxygen uptake during discrete and continuous submaximal running protocols and the repeatability of oxygen uptake on a day-to-day basis. The major findings of this study are presented in two sections: Study 2A and Study 2B. The major findings and recommendations from Study 2A are presented in Table 7.2. In this study, participants undertook discrete and continuous incremental submaximal exercise on the treadmill at identical speeds of 6.0, 7.0, 8.0 and 8.5 $km.hr^{-1}$.This study found that there was no difference in the oxygen uptake measured using either the discrete or continuous incremental submaximal protocols.

Table 7.2 Summary of major findings and recommendations from Study 2A

Summary of major findings	Recommendation
1. There was no difference in the oxygen uptake between the discrete and continuous submaximal protocols.	1. Continuous incremental submaximal exercise protocols may be acceptable for use with the pediatric population.
2. The heart rate of the participants was higher during the final two workloads of the continuous incremental submaximal exercise protocol.	2. Children exercising in a laboratory environment should be kept as cool as possible during the exercise test to minimise the effect of an increase in the thermal load on heart rate.
3. Accumulated oxygen deficit showed no differences when the discrete and continuous incremental submaximal protocols were compared.	3. Continuous incremental submaximal exercise protocols may make determining the AOD in the pediatric population less time-consuming.

The focus of section 2A of this thesis was to investigate whether the oxygen uptake during discrete and continuous incremental submaximal exercise was different. The first major finding was that the oxygen uptakes were not significantly different when the discrete and continuous incremental submaximal protocols were compared. The recommendation from this finding is that continuous incremental submaximal protocols, which require less time, may be appropriate for use with the pediatric population.

The second major finding of this study was that the heart rate was significantly higher in the final two workloads of the continuous incremental test when compared with the discrete test. Previous

research has suggested that continuous exercise causes an increase in the heat produced by the body (MacDougall et al. 1983; Rowland. 1996). The increase in heat causes the heart rate to increase. The recommendation from this finding is to endeavour to reduce the effects of heat on the participants by keeping them as cool as possible during exercise in the laboratory environment.

One of the recommendations from the first study of this thesis was to investigate less time-consuming protocols for determining the AOD in the pediatric population. The second study (Section 2A) investigated a less time-consuming method of determining the relationship between submaximal oxygen uptake and exercise intensity. There was no difference in AOD when either the discrete or continuous submaximal protocols was used to predict the supramaximal exercise intensity. Therefore, it is recommended that a continuous incremental submaximal exercise protocol may be appropriate to determine AOD in pediatric populations.

7.4 Summary and recommendations of Study 2B

Study 2B attempted to examine the methodology of the AOD technique by examining oxygen measurement repeatability. Specifically, Section 2B of this thesis investigated the repeatability of oxygen uptake during continuous incremental submaximal exercise over three days. A summary of the major findings and recommendations based on these findings is presented in Table 7.3. In this study, participants completed two continuous incremental submaximal exercise tests over three days. Each participant attended the laboratory at the same time on the first and the third day to minimise the effect of diurnal variation on oxygen uptake. This study found that the oxygen uptake during continuous incremental submaximal exercise was reliable.

Table 7.3 Summary of the major findings and recommendations from Study 2B

Summary of major findings	Recommendations
1. The oxygen uptake during continuous incremental submaximal exercise over three days is reliable.	1. That future studies investigate whether the repeatability of oxygen uptake varies over even greater time periods (7 days).
2. There was a large intraindividual variability in oxygen uptake over the testing period.	2. When using continuous incremental submaximal oxygen uptake to determine the relationship between submaximal oxygen uptake and exercise intensity individual linear regressions should be constructed. An assumed relationship will not account for the large individual variation observed in this population.
3. Greater variability of oxygen uptake at lower workloads.	3. Researchers investigating the anaerobic performance of children using AOD need to be aware that children exhibit a greater variability in oxygen uptake at lower submaximal exercise intensities.

The focus of this study was to investigate the repeatability of continuous incremental submaximal oxygen uptake during two testing sessions spread over three days. Previous researchers (Frost et al. 1995; Rogers et al. 1994; Unnithan et al. 1995) have found that oxygen uptake during discrete submaximal tests was reliable. This study found that oxygen uptake during continuous incremental submaximal exercise was reliable. A recommendation from this finding is that future

studies should investigate the repeatability of continuous incremental submaximal oxygen uptake over a greater time frame.

The second major finding of this study was that there was a large intraindividual variability in oxygen uptake during the testing period. This finding has major implications on the calculation of the relationship between submaximal oxygen uptake and exercise intensity. In the past, some researchers have used an assumed efficiency (Eriksson et al., 1973) in order to predict the supramaximal intensity. If an assumed efficiency is used, intraindividual variability will not be accounted for. The recommendation from this finding is that an individual relationship between submaximal oxygen uptake and exercise intensity should be constructed for every participant. Therefore individual variability in submaximal oxygen uptake will be accounted for.

A greater variability of oxygen uptake was observed at the lower submaximal exercise intensities. Sady et al. (1981; 1989) observed that this phenomenon was likely to occur in less mature children. This finding recommends that researchers investigating anaerobic performance in children using AOD should be aware that younger participants may exhibit a greater variability in oxygen uptake at the lower submaximal intensities and that this may affect the relationship between submaximal oxygen uptake and exercise intensity. Medbø (1991) proposed that submaximal intensities of 40 – 90% be used to determine the relationship between submaximal oxygen uptake and exercise intensity. Observations during these studies suggest that submaximal intensities of at least 50% be employed with the pediatric population.

7.5 Summary and recommendations of Study Three

Study Three of this thesis compared the AOD of pre-pubertal males using running and cycling exercise protocols. Table 7.4 presents a summary of the major findings and recommendations. Studies investigating anaerobic performance using AOD have traditionally used either cycling or running as the mode of exercise. Direct comparisons of AOD with running and cycling exercise have not been made. Study Three found that AOD was greater with running exercise when compared with cycling exercise.

Table 7.4 Summary of the major findings and recommendations from Study Three

Summary of major findings	Recommendations
1. That AOD was greater with running exercise when compared with cycling exercise.	1. That the size of the active muscle mass influences the size of AOD. Future studies could attempt to quantify the size of the muscle mass that is active during exercise.
2. Supramaximal test time, heart rate and RER were greater on the treadmill when compared with the cycle ergometer. The supramaximal workload on the cycle ergometer may have been too intense to completely exhaust the anaerobic system.	2. Cycling may not be the ideal mode of exercise for the pediatric population. Further investigations using cycle trained participants and untrained participants are warranted.
3. Strong Pearson correlation coefficient between Wingate Anaerobic Test Peak Power and AOD (ΣL) on the cycle ergometer.	3. Further investigations into the relationship between other measures of anaerobic performance such as the Wingate Anaerobic Test and Force-Velocity Test and AOD with the pediatric population.

The focus of this study was to investigate whether there was a difference in AOD with running and cycling exercise in the same group of participants. The study found that AOD was greater when running exercise was compared with cycling exercise in this sample of participants. It was suggested that the AOD might be greater with running exercise when compared with cycling exercise because the active muscle mass of running is greater than that of cycling. The recommendation based on this finding is for future studies to attempt to quantify the muscle mass that is active during exercise and correlate it with AOD.

The supramaximal test time, heart rate and RER was greater in the treadmill supramaximal exercise test when compared with the cycle ergometer supramaximal exercise test. Medbø and Tabata (1989) advocated that a supramaximal test time of between two and three minutes was optimal for determining AOD. The supramaximal test time for the treadmill was two minutes and six seconds, and the supramaximal test time for the cycle ergometer was one minute and one second. The supramaximal test time for the cycle ergometer was one minute shorter than the test time advocated by Medbø and Tabata (1989). The shorter test time could be a result of the intensity of the supramaximal workload, which may have been to high for the participants in this study to continue to exercise at. The recommendation based on this finding suggests that the cycling may not be the ideal mode of exercise for this population. Further studies investigating a comparison of running and cycling with participants trained in both modes of exercise or untrained participants are warranted.

The third major finding from this study was that there was a strong correlation between peak power (watts) generated during the Wingate Anaerobic Test and AOD ($\sum L$). The recommendation from this finding is that further investigations into the relationship between other measures of anaerobic performance such as the Wingate Anaerobic Test and Force-Velocity Test and AOD with pediatric populations.

7.6 General recommendations

The majority of the studies in this thesis have investigated the anaerobic performance of pre-pubertal males. Study One investigated the anaerobic performance of a cross-section of males that represented pre-pubertal, pubertal and post-pubertal development. The subsequent studies used pre-pubertal male participants only. The choice of participants for this thesis limits the findings and recommendations of this thesis to this population and largely neglects the responses of pubertal and post-pubertal males.

This thesis has not investigated the anaerobic performance of females using AOD, although the AOD of untrained pre-pubertal and trained pubertal females has been previously described (Carlson and Naughton, 1993; Naughton et al., 1997). Future studies investigating the anaerobic performance of females using AOD in both a cross-sectional and longitudinal design would provide some information on gender differences and responses to anaerobic exercise.

7.7 Issues of the use of accumulated oxygen deficit with children

The main issue with the use of AOD with the pediatric population remains with the determination of the relationship between submaximal oxygen uptake and exercise intensity. The participants in Studies Two and Three tolerated continuous incremental submaximal exercise protocols very well, but only four submaximal determinations could be made. This number falls well below the number suggested by Medbø et al. (1988) as desirable for determining the relationship between submaximal oxygen uptake and exercise intensity. Future investigations examining the use of two continuous incremental tests with three to four determinations may be more desirable.

7.8 Conclusion

The studies in this thesis have investigated the anaerobic performance of a cross-section of pre-pubertal, pubertal and post-pubertal males using AOD, alternative methods of determining the relationship between submaximal oxygen uptake and exercise intensity, the repeatability of oxygen uptake during continuous incremental submaximal exercise, and a comparison of AOD with running and cycling exercise. Together, the studies in this thesis have sought to explore the AOD as a means of examining the anaerobic performance of children. AOD has been shown to increase in a cross-section of children representing developmental stages from pre to post puberty, although the influence of training status in the cross-section of children studied could not be accounted for. A more time-efficient protocol for determining the relationship between submaximal oxygen uptake and exercise intensity using a continuous incremental submaximal protocol was also developed. AOD was also found to be greater with running when compared with cycling exercise. Future studies examining the anaerobic performance of children using AOD are warranted. These studies should focus on a longitudinal investigation of AOD in males and females and investigate whether AOD increases and is reflective of training in the pediatric population.

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APPENDICES

Appendix 1 Parent Letters

Appendix 1.1 Study One

Dear Parent's,

We are inviting your child to take part in a series of testing in the Exercise Physiology Laboratory at Victoria University of Technology (Footscray Campus). The purpose of this study is to investigate how much energy children use during very short but intense running tests on the treadmill.

Your child will be required to visit the laboratory seven times.

Visit 1: Familiarisation session (equipment and procedures explained).

Visit 2: A running test where children have the speed of the treadmill increased every minute until they volunteer their exhaustion.

Visit 3 - 5: In these tests, children will perform a series of six tests, each with a duration of six minutes. They will do between two and three tests per day allowing for at least 20 minutes rest between tests.

Visit 6- 7: The children will be challenged to run for as long as they can at one speed, which is harder than they have previously experienced.

Prior to the commencement of testing we will be asking your child to provide a sample of saliva. We are using the saliva to evaluate the amount of testosterone that your child has in their body. We will be using this value along with the Tanner Scale, (a diagrammatic scale of biological maturity), to establish where your child is on the maturity continuum.

During the final visit to the laboratory we will be asking your child to provide a small sample of blood from a vein in their arm. A trained and practiced physician performs this simple procedure. A surface anesthetic will be applied to the arm prior to the blood sample being taken. This procedure is routinely done in this laboratory, most recently on adolescent boys and girls. Research into this procedure has found that children can benefit from this procedure if they are faced with giving blood as part of a medical diagnosis.

Having conducted these tests on children of all ages, over the past year, we can assure you that they really enjoy the tasks and perform them extremely well. The children also benefit from visits to the laboratory. They find the experiences interesting and appear to learn a great deal about their bodies' response to exercise. Throughout the tests consistent positive reinforcement is given to encourage the children to do their best. All children and parents should be aware that their child is free to withdraw from the study at any time.

For all testing, shorts and running shoes are preferred. Following each testing session your child will receive a juice and a health bar.

If you require transport in order to enable your child to participate please do not hesitate to contact me to arrange it.

We thank you for your interest in improving our understanding of the nature of the child's response to exercise.

Your cooperation is valued.

Kathryn Meldrum.
Professor John Carlson.

Appendix 1.2 Study 2A

Dear Parent's,

We are inviting your child to take part in a series of testing in the Exercise Physiology Laboratory at Victoria University of Technology (Footscray Campus). The purpose of this study is to investigate how much energy children use during very short but intense running tests on the treadmill.

Your child will be required to visit the laboratory seven times.

Visit 1:	Familiarisation session (equipment and procedures explained).
Visit 2:	A running test where children have the speed of the treadmill increased every minute until they volunteer their exhaustion.
Visit 3 - 4:	In these tests, children will perform a series of six tests, each with a duration of four minutes. They will do between two and three tests per day allowing for at least 20 minutes rest between tests.
Visit 5:	Continuous treadmill running test at submaximal speeds.
Visit 6- 7:	The children will be challenged to run for as long as they can at one speed, which is harder than they have previously experienced.

During testing we will be asking you to identify where your child is on the Tanner Scale, (a diagrammatic scale of biological maturity), to establish where your child is on the maturity continuum. You will be receiving a separate letter about this through the mail.

Having conducted these tests on children of all ages, over the past year, we can assure you that they really enjoy the tasks and perform them extremely well. The children also benefit from visits to the laboratory. They find the experiences interesting and appear to learn a great deal about their bodies' response to exercise. Throughout the tests consistent positive reinforcement is given to encourage the children to do their best. All children and parents should be aware that their child is free to withdraw from the study at any time.

For all testing, shorts and running shoes are preferred. Following each testing session your child will receive a juice and a health bar.

If you require transport in order to enable your child to participate please do not hesitate to contact me to arrange it.

We thank you for your interest in improving our understanding of the nature of the child's response to exercise.

Your cooperation is valued.

Kathryn Meldrum.
Professor John Carlson.

Appendix 1.3 Parent Letter Study 2B

Dear Parent's,

We are inviting your child to take part in a series of testing in the Exercise Physiology Laboratory at Victoria University of Technology (Footscray Campus). The purpose of this study is to investigate whether children's oxygen use during exercise varies over two testing sessions one day apart.

Your child will be required to visit the laboratory three times.

Visit 1: Familiarisation session (equipment and procedures explained).

Visit 2: Continuous treadmill running test at submaximal speeds **OR** Cycling on cycle ergometer

Visit 3 The same test as visit 2.

During testing we will be asking you to identify where your child is on the Tanner Scale, (a diagrammatic scale of biological maturity), to establish where your child is on the maturity continuum. You will be receiving a separate letter about this through the mail.

Having conducted these tests on children of all ages, over the past year, we can assure you that they really enjoy the tasks and perform them extremely well. The children also benefit from visits to the laboratory. They find the experiences interesting and appear to learn a great deal about their bodies' response to exercise. Throughout the tests consistent positive reinforcement is given to encourage the children to do their best. All children and parents should be aware that their child is free to withdraw from the study at any time.

For all testing, shorts and running shoes are preferred. Following each testing session your child will receive a juice and a health bar.

If you require transport in order to enable your child to participate please do not hesitate to contact me to arrange it.

We thank you for your interest in improving our understanding of the nature of the child's response to exercise.

Your cooperation is valued.

Kathryn Meldrum.
Professor John Carlson.

Appendix 1.4 Parent Letter Study Three

Dear Parent's,

We are inviting your child to take part in a series of testing in the Exercise Physiology Laboratory at Victoria University of Technology (Footscray Campus). The purpose of this study is to investigate how much energy children use during very short but intense tests running on the treadmill and cycling on a bicycle ergometer.

Your child will be required to visit the laboratory six times.

Visit 1: Familiarisation session (equipment and procedures explained).

Visit 2 - 3: During visits two and three either two running or two cycling tests will be performed. The order of the cycling and running tests is random.
The first cycling test will be a continuous cycling test at easy resistances. Then after a rest a cycling ergometer test where children have the resistance of the bicycle ergometer increased every minute until they volunteer their exhaustion.

The first running test will be a continuous running test at easy speeds. Then after a rest a treadmill test where the children will have the speed of the treadmill increased every minute until they volunteer their exhaustion.

Visit 4 - 5: The children will be challenged to run or cycle for as long as they can at one speed, which is harder than they have previously experienced.

Visit 6 The children will be asked to ride on the cycle ergometer as fast as they can for 30 seconds.

During testing we will be asking you to identify where your child is on the Tanner Scale, (a diagrammatic scale of biological maturity), to establish where your child is on the maturity continuum. You will be receiving a separate letter about this through the mail.

Having conducted these tests on children of all ages, over the past year, we can assure you that they really enjoy the tasks and perform them extremely well. The children also benefit from visits to the laboratory. They find the experiences interesting and appear to learn a great deal about their bodies' response to exercise. Throughout the tests consistent positive reinforcement is given to encourage the children to do their best. All children and parents should be aware that their child is free to withdraw from the study at any time.

For all testing, shorts and running shoes are preferred. Following each testing session your child will receive a juice and a health bar.

If you require transport in order to enable your child to participate please do not hesitate to contact me to arrange it.

We thank you for your interest in improving our understanding of the nature of the child's response to exercise.

Your cooperation is valued.

Kathryn Meldrum.
Professor John Carlson.

Appendix 1.5 Separate parent letter to explain the Tanner Scale diagrams

Pediatric Exercise Research Unit
Victoria University of Technology
Ballarat Road
Footscray.

Dear Parent's,

Thank you for your interest and for allowing your son to participate in my research. I intend to start testing your son on the 4th. of April. Please make sure that he brings shoes suitable for running in and shorts and a t - shirt.

For the purposes of research I need to know your son's stage of pubertal development. This can be done with your help. I have enclosed one page of pictures called the Tanner Scale. Doctor's use this scale to identify the developmental stages of children. These stages are numbered from one to five. Please circle the stage of development that you believe your son to be at and return it to me in the pre - paid envelope enclosed. Please print your son's name on the top of the page.

Thank you for your assistance.

Kathryn Meldrum.

Parent Consent Form

<u>This form is to accompany the Medical Report Form before your child becomes involved in the program.</u>

I, ...
(your name)

Of,
.....
.....
.....
(your address)

give consent for my Son/Daughter..... to be
(child's name)
involved in the program with Victoria University of Technology.

Signed:.....
Date:.....

Appendix 1.7 Parent medical consent form.

CONFIDENTIAL MEDICAL REPORT

PLEASE COMPLETE AND RETURN AS SOON AS POSSIBLE

Note: **Both this form and the Parental Consent Form must be completed** for your child to secure a place on this program.

Name of Child:.....

Name of Parent/Guardian:.....

Address: .....
.....

Contact number: Day.....Evening:.....

Medical Information:

1. Medicare Number: .....
2. Ambulance Number:.....
3. Private Health Insurance YES\NO
   IF YES, Please Name:.....
4. Last Tetanus Immunisation was?.....
5. Does your son/daughter suffer from any regular complaints or allergies? E. g. Migraine, asthma. Please be as specific and as detailed as possible, and include instructions for dealing with the situation should it arise. E.g. Tablets etc.

.....
.....
.....
.....

In case of accident, or the need for medical assistance, please give your consent to take your Son/Daughter to the appropriate agency (Hospital/Clinic), by signing this form.

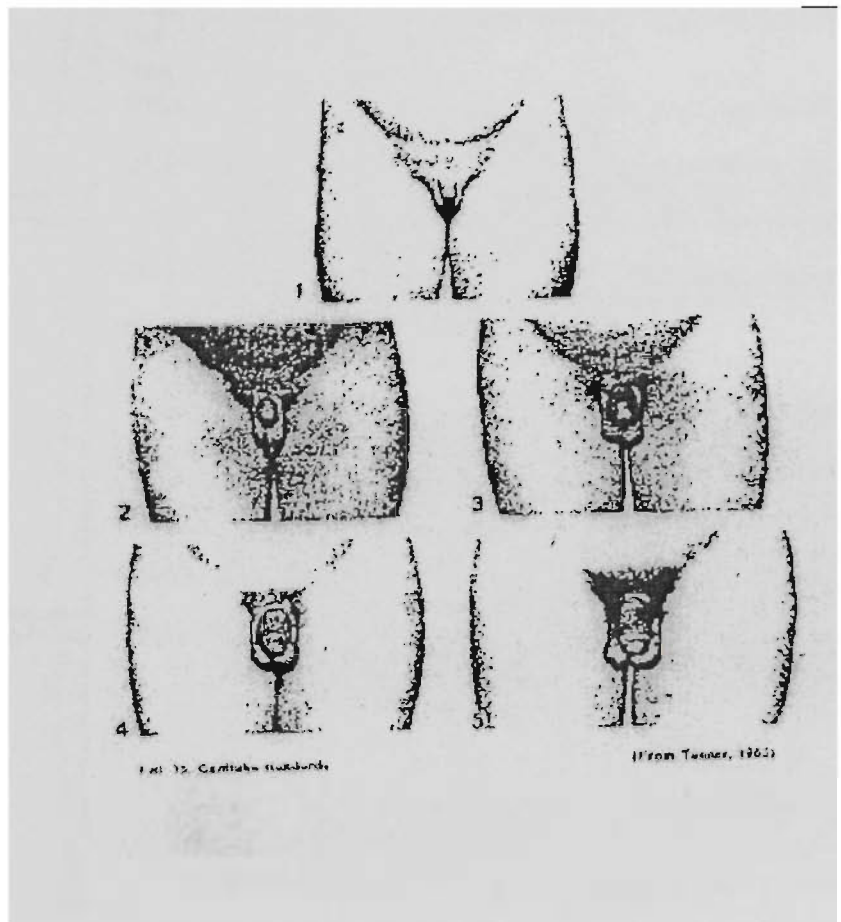
Thank you for your assistance.

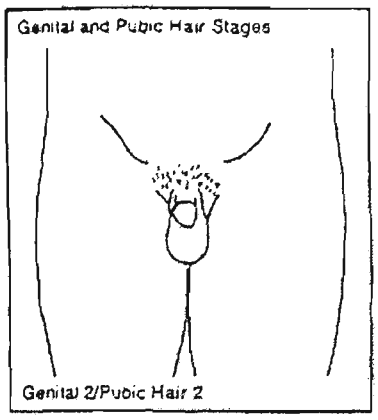
I,....., give my consent for Kathryn Meldrum and Professor John Carlson to administer or seek the appropriate medical attention for my Son/Daughter.....(Insert child's name) during the testing at Victoria University of Technology (Footscray Campus) and agree to cover any costs incurred.

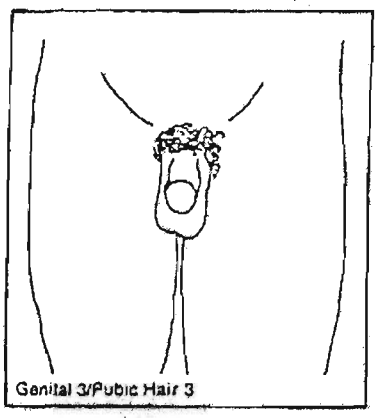
Signed:.....Print Name:.....
Date:.....

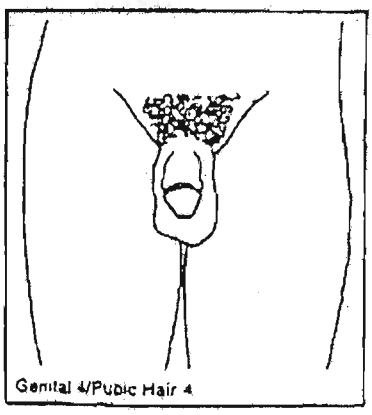
Appendix 2 Tanner scale and stretching exercises

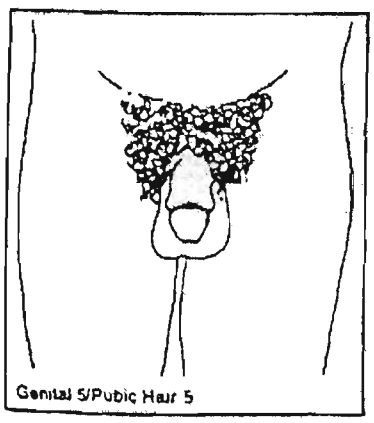
Appendix 2.1 Tanner scale diagrams used in Study One











STAGES OF PUBERTY

Ages of attainment of successive stages of pubertal sexual development are given in the height centile chart overpage. The stage Pubic Hair 2+ represents the state of a child who shows the pubic hair appearance stage 2 but not stage 3 (see below). The centiles for age at which this state is normally seen are given, the 97th centile being considered as the early limit, the 3rd centile as the late limit. The child's puberty stages may be plotted at successive ages (Tanner, *Growth at Adolescence*, 2nd Ed., 1962). Testis sizes are judged by comparison with the Prader orchidometer (Zachmann, Prader, Kind, Hallinger and Budliger, *Helv. Paed. Acta.* 29, 61–72, 1974).

Genital (penis) development:

Stage 1. Pre-adolescent, testes, scrotum and penis are of about the same size and proportion as in early childhood.

Stage 2. Enlargement of scrotum and testes. Skin of scrotum reddens and changes in texture. Little or no enlargement of penis at this stage.

Stage 3. Enlargement of the penis which occurs at first mainly in length. Further growth of the testes and scrotum.

Stage 4. Increased size of penis with growth in breadth and development of glans. Testes and scrotum larger; scrotal skin darkened.

Stage 5. Genitalia adult in size and shape.

Pubic hair:

Stage 1. Pre-adolescent. The vellus over the pubes is not further developed than that over the abdominal wall, i.e. no pubic hair.

Stage 2. Sparse growth of long, slightly pigmented downy hair, straight or slightly curled at the base of the penis.

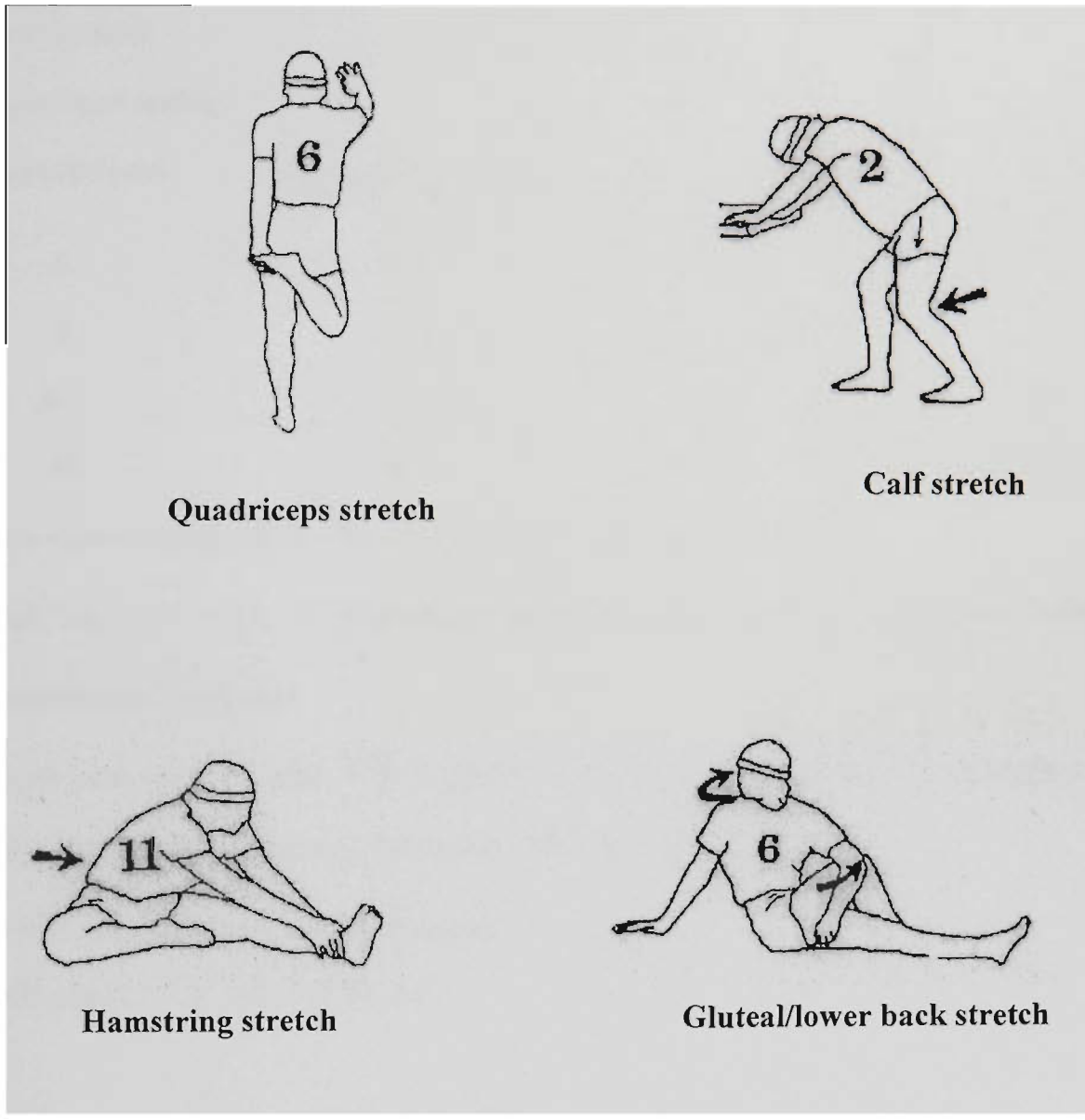
Stage 3. Considerably darker, coarser and more curled. The hair spreads sparsely over the junction of the pubes.

Stage 4. Hair now adult in type, but area covered is still considerably smaller than in the adult. No spread to the medial surface of thighs.

Stage 5. Adult in quantity and type with distribution of the horizontal (or classically 'feminine') pattern. Spread to medial surface of thighs but not up linea alba or elsewhere above the base of the inverse triangle (spread up linea alba occurs late and is rated stage 6).

Tanner Scale representations. The Adelaide Children's Hospital, 1988.
Used in Studies Two, Three and Four.

Appendix 2.3 Example of stretching exercises prior to the supramaximal test



Anderson, 1980.

Appendix 3 An example of the calculation of the supramaximal workload.

"Peter" aged 12.5 years has a mass of 30.04 kg and his peak $\dot{V}O_2$ was 58.87 $ml^{-1}.kg^{-1}.min^{-1}$ on a motorised treadmill.

3. Submaximal testing:

Treadmill Speed	$\dot{V}O_2$ $mL^{-1}.kg^{-1}.min^{-1}$
6	33.04
8	45.36
9.1	51.66
10	56.77

The linear regression equation [y = a + b(x)] computed from the above data is:

$\dot{V}O_2$ ($mL^{-1}.kg^{-1}.min^{-1}$) = 0.41 + 5.05(treadmill speed) which has a correlation coefficient of 0.99

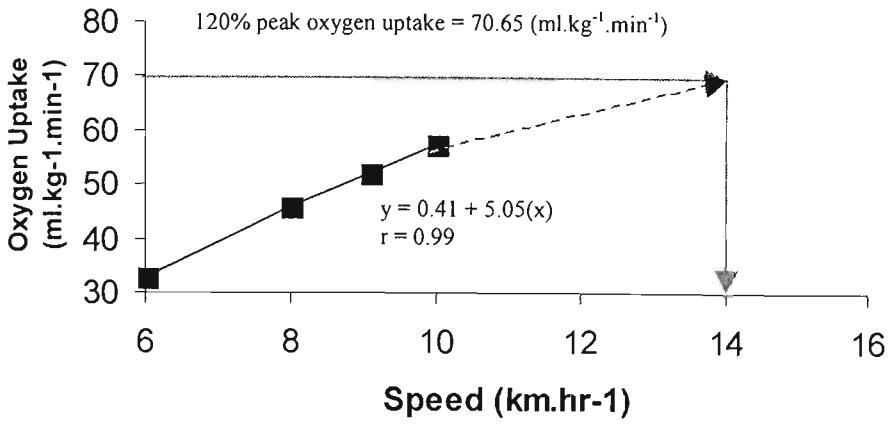
4. Supramaximal prediction

The oxygen uptake at 120% peak $\dot{V}O_2$ is (58.87 x 1.2) = 70.64 $mL^{-1}.kg^{-1}.min^{-1}$. From the linear regression the workload representing 120% peak $\dot{V}O_2$ is calculated.

70.64 = 0.41 + 5.05 (unknown treadmill speed)

Treadmill speed (x) = y – a/b = 13.9 $km.hr^{-1}$

Sample calculation from raw data



Appendix 4 Statistics from Study One

Appendix 4.1 Descriptive characteristics and maximal effort profiles

Factorial Analysis

Condition = Category (Pre-pubertal, pubertal and post-pubertal)

Analysis of Variance

Variable Age
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	167.213	83.606	52.834	0.000
Within Groups	27	42.726	1.582		
Total	29	209.939			

Variable Mass
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	7142.931	3571.465	62.568	0.000
Within Groups	27	1541.208	57.082		
Total	29	8684.138			

Variable Height
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	5976.914	2988.457	43.211	0.000
Within Groups	27	1867.301	69.159		
Total	29	7844.215			

Variable: Peak oxygen uptake ($L.min^{-1}$)
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	43.405	21.703	71.727	0.000
Within Groups	27	8.169	0.303		
Total	29	51.574			

Variable: Peak oxygen uptake ($mL.kg^{-1}.min^{-1}$)
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	476.699	238.350	4.457	0.021
Within Groups	27	1443.961	53.480		
Total	29	1920.660			

Variable: Peak heart rate
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	306.067	153.033	2.778	0.080
Within Groups	27	1487.300	55.085		
Total	29	1793.367			

Appendix 4.2 Tanner Scale and salivary testosterone profiles

Factorial Analysis

Condition = Category (Pre-pubertal, pubertal and post-pubertal)

Analysis of Variance

Variable Tanner Scale
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	80.600	40.3000	107.733	0.000
Within Groups	27	10.100	0.374		
Total	29	90.700			

Variable Salivary testosterone
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	423760.343	211880.172	33.291	0.000
Within Groups	27	165477.096	6364.504		
Total	29	589237.439			

Appendix 4.3 Mean lowest and highest submaximal exercise intensities

Factorial Analysis

Condition = Category (Pre-pubertal, pubertal and post-pubertal)

Analysis of Variance

Variable — Mean lowest submaximal intensity
By Variable — Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	57.387	28.693	0.262	0.771
Within Groups	27	2624.800	109.367		
Total	29	2682.187			

Variable — Mean highest submaximal intensity
By Variable — Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	328.183	164.092	3.097	0.064
Within Groups	27	1271.686	52.987		
Total	29	1599.870			

Appendix 4.4 Submaximal oxygen uptake and exercise intensity regression equations

Factorial Analysis

Condition = Category (Pre-pubertal, pubertal and post-pubertal)

Analysis of Variance

Variable Y intercept
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	36.717	18.358	3.051	0.065
Within Groups	27	150.433	6.017		
Total	29	187.150			

Variable Slope
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	0.000	0.000	0.372	0.693
Within Groups	27	0.171	0.000		
Total	29	0.176			

Variable Pearson correlation (r)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	0.001	0.000	2.439	0.108
Within Groups	27	0.009	0.000		
Total	29	0.108			

Appendix 4.5 Supramaximal treadmill speeds

Factorial Analysis

Condition = Category (Pre-pubertal, pubertal and post-pubertal)

Analysis of Variance

Variable Treadmill speed
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	97.437	48.719	7.877	0.002
Within Groups	27	154.621	6.185		
Total	29	252.058			

Appendix 4.6 Accumulated oxygen deficits and supramaximal performance parameters

Factorial Analysis

Condition = Pre-pubertal and pubertal

Analysis of Variance

Variable Accumulated Oxygen Deficit (ΣL)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	10.59	10.59	10.59	0.005
Within Groups	18	16.99	0.999		
Total	19	28.58			

Variable Accumulated Oxygen Deficit ($\Sigma mL.kg^{-1}$)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	2299.95	2299.95	6.88	0.018
Within Groups	18	5677.33	333.96		
Total	19	7977.28			

Variable Supramaximal Heart Rate
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	7.73	7.73	0.141	0.712
Within Groups	18	1151.34	54.82		
Total	19	1855.06			

Variable Supramaximal Test Time
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	3357.93	3357.93	0.470	0.502
Within Groups	18	121564.5	7150.85		
Total	19	124922.5			

Variable RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.006	0.006	0.944	0.345
Within Groups	18	0.121	0.007		
Total	19	0.128			

Factorial Analysis

Condition = Pre-pubertal and post-pubertal

Analysis of Variance

Variable Accumulated Oxygen Deficit (ΣL)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	29.75	29.75	24.46	0.000
Within Groups	18	19.45	1.21		
Total	19	49.20			

Variable Accumulated Oxygen Deficit ($\Sigma mL.kg^{-1}$)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	757.50	757.50	2.72	0.118
Within Groups	18	4441.89	277.61		
Total	19	5199.40			

Variable Supramaximal Heart Rate
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	241.73	241.73	3.35	0.086
Within Groups	18	1151.37	71.96		
Total	19	1393.11			

Variable Supramaximal Test Time
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	3534.65	3534.65	0.927	0.350
Within Groups	18	61029.5	3814.34		
Total	19	64564.2			

Variable RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.001	0.001	2.83	0.112
Within Groups	18	0.008	0.0005		
Total	19	0.105			

Factorial Analysis

Condition = Pubertal and post-pubertal

Analysis of Variance

Variable Accumulated Oxygen Deficit (ΣL)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	5.04	5.04	2.66	0.123
Within Groups	18	28.41	1.89		
Total	19	33.46			

Variable Accumulated Oxygen Deficit ($\Sigma mL.kg^{-1}.min^{-1}$)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	341.52	341.52	0.802	0.385
Within Groups	18	6385.25	425.68		
Total	19	6726.78			

Variable Supramaximal Heart Rate
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	317.09	317.09	4.39	0.053
Within Groups	18	1082.43	72.16		
Total	19	1399.52			

Variable Supramaximal Test Time
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	12730.88	12730.88	1.59	0.226
Within Groups	18	119839.1	7989.27		
Total	19	132570.0			

Variable RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.002	0.002	0.386	0.553
Within Groups	18	0.08	0.005		
Total	19	0.008			

ANCOVA

Variable	AOD (L)
By Variable	Condition
With	Mass

Source of Variation	Mean D.F.	Sum Squares	Mean Squares	F Ratio	F Prob.
Covariates	1	2.201	2.201	0.209	
Mass	1	2.201	2.201	0.209	
Main effects	2	3.072	1.536	0.329	
CAT	2	3.072	1.536	0.329	
Explained	3	32.668	10.889	8.285	0.001
Residual	23	30.230	1.314		
Total	26	62.898	2.419		

A: Predictors (constant) Mass
B: Dependent variable AOD(L)

Appendix 4.7 Effect sizes for Study One

	Preadolescent/ Adolescent	Adolescent/ Post adolsecent	Pre adolescent/ Post adolescent
Peak oxygen uptake ($L.min^{-1}$)	1.70	3.31	5.39
Peak oxygen uptake ($mL.kg.min^{-1}$)	0.39	0.93	1.18
Salivary testosterone (pmol)	1.83	1.64	4.71
AOD (L)	1.54	0.92	2.42
AOD ($mL.kg^{-1}$)	1.42	0.3	0.91

Appendix 4.8 pH pre and post-supramaximal exercise

Factorial Analysis

Condition = Pre-pubertal and pubertal

Analysis of Variance

Variable: Plasma pH at Rest
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.001	0.001	0.845	0.385
Within Groups	8	0.02	0.01		
Total	9	0.01			

Variable: Plasma pH at minute one post exercise
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.001	0.003	0.214	0.656
Within Groups	8	0.03			
Total	9	0.03			

Variable: Plasma pH at minute three post exercise
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.01	0.01	0.311	0.593
Within Groups	8	0.02	0.003		
Total	9	0.03			

Variable Plasma pH at minute five post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.001	0.001	0.343	0.574
Within Groups	8	0.02	0.002		
Total	9	0.02			

Variable Plasma pH at minute seven post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.003	0.003	0.584	0.467
Within Groups	8	0.02	0.002		
Total	9	0.02			

Variable Plasma pH at minute ten post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.000004	0.000004	0.002	0.965
Within Groups	8	0.01	0.002		
Total	9	0.01			

Variable Plasma pH at minute fifteen post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.003	0.003	1.60	0.241
Within Groups	8	0.01	0.002		
Total	9	0.01			

Variable	Plasma pH at minute twenty post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.00005	0.00005	0.067	.80.3
Within Groups	8	0.006	0.0007		
Total	9	0.006			

Factorial Analysis

Condition = Pre-pubertal and post-pubertal

Analysis of Variance

Variable	Plasma pH at Rest
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.01	0.01	0.060	0.812
Within Groups	8	2.12	0.266		
Total	9	2.14			

Variable	Plasma pH at minute one post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	21.31	21.31	4.57	0.065
Within Groups	8	37.28	4.66		
Total	9	58.59			

Variable Plasma pH at minute three post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	40.00	40.00	10.39	0.012
Within Groups	8	30.80	3.85		
Total	9	70.80			

Variable Plasma pH at minute five post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	37.24	37.24	7.60	0.025
Within Groups	8	39.17	4.89		
Total	9	76.42			

Variable Plasma pH at minute seven post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	35.34	35.34	6.63	0.033
Within Groups	8	42.59	5.32		
Total	9	77.93			

Variable Plasma pH at minute ten post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	29.92	29.92	6.13	0.038
Within Groups	8	39.01	4.87		
Total	9	68.94			

Variable Plasma pH at minute fifteen post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	16.90	16.90	4.11	0.077
Within Groups	8	32.89	4.11		
Total	9	49.79			

Variable Plasma pH at minute twenty post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	28.56	28.56	6.95	0.030
Within Groups	8	32.84	4.10		
Total	9	49.79			

Factorial Analysis

Condition = Pubertal and post-pubertal

Analysis of Variance

Variable Plasma pH at Rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.003	0.003	3.73	0.090
Within Groups	8	0.007	0.0009		
Total	9	0.01			

Variable Plasma pH at minute one post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.007	0.007	1.24	0.297
Within Groups	8	0.04	0.005		
Total	9	0.05			

Variable	Plasma pH at minute three post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.007	0.007	1.08	0.328
Within Groups	8	0.05	0.006		
Total	9	0.06			

Variable	Plasma pH at minute five post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.008	0.008	1.32	0.283
Within Groups	8	0.05	0.006		
Total	9	0.05			

Variable	Plasma pH at minute seven post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.009	0.009	1.72	0.225
Within Groups	8	0.007	0.005		
Total	9	0.01			

Variable	Plasma pH at minute ten post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.01	0.01	2.30	0.168
Within Groups	8	0.03	0.004		
Total	9	0.04			

Variable	Plasma pH at minute fifteen post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.00009	0.00009	0.037	0.853
Within Groups	8	0.02	0.002		
Total	9	0.02			

Variable	Plasma pH at minute twenty post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.0006	0.0006	0.527	0.489
Within Groups	8	0.01	0.001		
Total	9	0.01			

Appendix 4.8.1 Scheffès method for between participant effects

F value for pH

Time (min)	Pre-pubertal	Pubertal	Post-pubertal
Rest - Post 1	152.21*	11.74*	134.29*
Post 1 - Post 3	4.73	1.07	0.60
Post 3 - Post 5	0.00	1.39	3.33
Post 5 - Post 7	42.66*	0.58	2.25
Post 7 - Post 10	2.25	42.66*	42.25*
Post 10 - Post 15	60.16*	0.00	10.01*
Post 15 - Post 20	1.42	6.76	75.25*

* Significantly different ($P < 0.05$)

Appendix 4.9 Blood lactate pre and post-supramaximal exercise

Factorial Analysis

Condition = Pre-pubertal and pubertal

Analysis of Variance

Variable Blood lactate at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.289	0.289	2.65	0.142
Within Groups	8	12.68	0.109		
Total	9	15.82			

Variable Blood lactate at minute one post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.900	0.900	0.688	0.431
Within Groups	8	10.46	1.30		
Total	9	11.36			

Variable Blood lactate at minute three post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	2.70	2.70	2.09	0.186
Within Groups	8	10.35	1.29		
Total	9	13.05			

Variable Blood lactate at minute five post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	2.30	2.30	1.77	0.220
Within Groups	8	10.41	1.30		
Total	9	12.71			

Variable Blood lactate at minute seven post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	3.13	3.13	1.97	0.197
Within Groups	8	12.68	1.58		
Total	9	15.82			

Variable Blood lactate at minute ten post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1.76	1.76	1.18	0.308
Within Groups	8	11.93	1.49		
Total	9	13.69			

Variable Blood lactate at minute fifteen post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1.02	1.02	0.94	0.359
Within Groups	8	8.65	1.08		
Total	9	9.68			

Variable Blood lactate at minute twenty post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	3.13	3.13	2.75	0.135
Within Groups	8	9.10	1.13		
Total	9	12.23			

Factorial Analysis

Condition = Pre-pubertal and post-pubertal

Analysis of Variance

Variable Blood lactate at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.01	0.01	0.06	0.812
Within Groups	8	2.12	0.26		
Total	9	2.14			

Variable Blood lactate at minute one post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	21.31	21.31	4.57	0.065
Within Groups	8	37.28	4.66		
Total	9	58.59			

Variable Blood lactate at minute three post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	40.00	40.00	10.39	0.012
Within Groups	8	30.08	3.85		
Total	9	70.80			

Variable Blood lactate at minute five post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	37.24	37.24	7.60	0.025
Within Groups	8	39.17	4.89		
Total	9	76.42			

Variable Blood lactate at minute seven post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	35.34	35.34	6.63	0.033
Within Groups	8	42.59	5.32		
Total	9	77.93			

Variable Blood lactate at minute ten post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	29.92	29.92	6.13	0.038
Within Groups	8	39.01	4.87		
Total	9	68.94			

Variable Blood lactate pH at minute fifteen post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	16.90	16.90	4.11	0.077
Within Groups	8	32.89	4.11		
Total	9	49.79			

Variable Blood lactate at minute twenty post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	28.56	28.56	6.95	0.030
Within Groups	8	32.84	4.10		
Total	9	61.40			

Factorial Analysis

Condition = Pubertal and post-pubertal

Analysis of Variance

Variable Blood lactate at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.44	0.44	1.31	0.284
Within Groups	8	2.68	0.33		
Total	9	3.12			

Variable Blood lactate at minute one post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	30.97	30.97	6.50	0.034
Within Groups	8	38.08	4.76		
Total	9	69.05			

Variable Blood lactate at minute three post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	21.90	21.90	4.46	0.068
Within Groups	8	39.23	4.90		
Total	9	61.13			

Variable Blood lactate at minute five post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	21.02	21.02	3.51	0.098
Within Groups	8	47.80	5.97		
Total	9	68.82			

Variable Blood lactate at minute seven post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	17.42	17.42	2.69	0.139
Within Groups	8	51.76	6.47		
Total	9	68.18			

Variable Blood lactate at minute ten post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	17.16	17.16	2.77	0.135
Within Groups	8	49.54	6.190		
Total	9	66.70			

Variable Blood lactate at minute fifteen post exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	9.60	9.60	1.87	0.208
Within Groups	8	40.97	5.12		
Total	9	50.58			

Variable	Blood lactate at minute twenty post exercise
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	12.76	12.76	2.46	0.155
Within Groups	8	41.44	5.18		
Total	9	54.20			

Appendix 4.9.1 Scheffè's method for between participant effects

F value for Blood lactate

Time (min)	Pre-pubertal	Pubertal	Post-pubertal
Rest - Post 1	84.66*	130.21*	54.11*
Post 1 - Post 3	5.86	2.79	0.04
Post 3 - Post 5	42.66*	4.23	0.29
Post 5 - Post 7	11.66*	12.25*	9.22*
Post 7 - Post 10	1.55	25.13*	35.00*
Post 10 - Post 15	0.56	1.75	25.01*
Post 15 - Post 20	55.80*	11.55	27.93*

* Significantly different ($P < 0.05$)

Appendix 4.10 Haemoglobin concentration pre supramaximal exercise

Factorial Analysis

Condition = Pre-pubertal and pubertal

Analysis of Variance

Variable Haemoglobin concentration at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	5.59	5.59	4.05	0.079
Within Groups	8	11.03	1.37		
Total	9	16.62			

Factorial Analysis

Condition = Pre-pubertal and post-pubertal

Analysis of Variance

Variable Haemoglobin concentration at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	12.32	12.32	49.28	0.000
Within Groups	8	2.00	0.250		
Total	9	14.32			

Factorial Analysis

Condition = Pubertal and post-pubertal

Analysis of Variance

Variable Haemoglobin concentration at rest
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1.31	1.31	1.04	0.337
Within Groups	8	10.04	1.25		
Total	9	11.35			

Appendix 5 Statistics Study Two

Appendix 5.1 Submaximal oxygen uptake test parameters

Factorial Analysis

Condition = Continuous and discrete submaximal protocol

Analysis of Variance

Variable: Oxygen uptake at 6 km.hr.$^{-1}$
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.116	0.116	.015	0.903
Within Groups	18	135.303	7.517		
Total	19	135.418			

Variable: Oxygen uptake at 7 km.hr.$^{-1}$
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	102.876	102.876	1.383	0.255
Within Groups	18	1339.281	74.404		
Total	19	1442.157			

Variable: Oxygen uptake at 8 km.hr.$^{-1}$
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	23.814	23.814	1.604	0.226
Within Groups	18	207.842	14.846		
Total	19	231.656			

Variable	Oxygen uptake at 8.5 km.hr.$^{-1}$
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	30.085	30.085	4.148	0.61
Within Groups	18	101.532	7.252	4.148	
Total	19	131.617			

Factorial Analysis

Condition = Continuous and discrete submaximal protocol

Analysis of Variance

Variable	Ventilation at 8.5 km.hr^{-1} in the second minute
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	225.836	225.836	8.495	0.010
Within Groups	18	398.759	26.584		
Total	19	624.595			

Variable	Ventilation at 8.5 km.hr^{-1} in the third minute
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	203.505	203.505	6.998	0.018
Within Groups	18	436.165	29.077		
Total	19	639.671			

Variable RER at 8 km.hr^{-1} in the second minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.013	0.013	6.424	0.020
Within Groups	18	0.037	0.002		
Total	19	0.051			

Variable RER at 8 km.hr^{-1} in the third minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.003	0.003	1.215	0.284
Within Groups	18	0.050	0.002		
Total	19	0.053			

Variable RER at 8 5km.hr^{-1} in the second minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.016	0.016	13.856	0.002
Within Groups	18	0.018	0.001		
Total	19	0.035			

Variable RER at 8 5km.hr^{-1} in the third minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.011	0.011	8.951	0.009
Within Groups	18	0.019	0.001		
Total	19	0.030			

Variable Heart rate at 8 km.hr^{-1} in the first minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1022.450	1022.450	8.854	0.008
Within Groups	18	2078.50	115.47		
Total	19	3100.950			

Variable Heart rate at 8 km.hr^{-1} in the second minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	661.250	661.250	7.285	0.014
Within Groups	18	1663.700	90.761		
Total	19	2294.950			

Variable Heart rate at 8 km.hr^{-1} in the third minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	460.800	460.800	4.843	0.041
Within Groups	18	1712.40	95.133		
Total	19	2173.200			

Variable Heart rate at 8 5km.hr^{-1} in the first minute
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	2738.000	2738.000	27.552	0.000
Within Groups	18	1590.000	99.375		
Total	19	4328.000			

Variable	Heart rate at 8.5 $km.hr^{-1}$ in the second minute
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1720.888	1720.888	19.212	0.000
Within Groups	18	1433.111	89.569		
Total	19	3154.000			

Variable	Heart rate at 8.5 $km.hr^{-1}$ in the third minute
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	1250.000	1250.000	12.500	0.002
Within Groups	18	1600.00	100.000		
Total	19	2850.00			

Appendix 5.2 Oxygen uptake and heart rate test one and test two

Oxygen uptake test one and test two

Paired Samples T Test

Pair	D.F.	t	Sig. (2-tailed)
Workload One	11	0.182	0.859
Workload Two	11	-0.773	0.456
Workload Three	11	-0.880	0.398

Heart rate test one and test two

Pair	D.F.	t	Sig. (2-tailed)
Workload One	11	-1.943	0.078
Workload Two	11	-0.860	0.408
Workload Three	11	-0.634	0.539

Paired Samples Correlations

Oxygen uptake test one and two

Pair	N	Correlation	Significance
Workload One	12	0.933	0.000
Workload Two	12	0.849	0.000
Workload Three	12	0.885	0.000

Heart rate test one and test two

Pair	N	Correlation	Significance
Workload One	12	0.755	0.005
Workload Two	12	0.859	0.000
Workload Three	12	0.905	0.000

Appendix 5.3 Mean lowest and highest submaximal intensity

Factorial Analysis

Condition = Continuous and discrete submaximal protocol

Analysis of Variance

Variable Lowest intensity of submaximal exercise
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.420	0.420	0.019	0.891
Within Groups	18	394.125	21.896		
Total	19	394.564			

Variable Mean highest submaximal intensity
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	132.098	132.098	2.658	0.120
Within Groups	18	894.414	49.690		
Total	19	1026.512			

Appendix 5.4 Comparison of heart rate and oxygen uptake over three testing sessions.

Factorial Analysis

Condition = Three separate testing sessions

Analysis of Variance

Variable: Oxygen uptake ($mL.kg^{-1}.min^{-1}$)
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	12.669	6.334	0.685	0.513
Within Groups	27	249.725	9.249		
Total	29	262.394			

Variable: Heart rate
By Variable: Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	2	408.642	204.321	1.365	0.273
Within Groups	27	4042.301	149.715		
Total	29	4450.943			

Appendix 5.5 Mean linear regression equation and supramaximal treadmill speed

Factorial Analysis

Condition = Continuous and discrete submaximal protocol

Analysis of Variance

Variable Slope
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	2.11	2.11	2.24	0.151
Within Groups	18	16.95	0.94		
Total	19	19.07			

Variable Y-intercept
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.002	0.002	3.83	0.066
Within Groups	18	0.01	0.0005		
Total	19	0.01			

Variable Correlation coefficient (r)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	0.00008	0.00008	0.40	0.532
Within Groups	18	0.003	0.0001		
Total	19	0.003			

Variable	Speed
By Variable	Condition

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F Prob.
Between Groups	1	3.63	3.63	1.46	0.242
Within Groups	18	44.81	2.49		
Total	19	48.45			

Appendix 5.6 Linear regression equation parameters test one and test two

Paired samples T test

Linear regression equation parameters test one and test two

Pair	D.F.	t	Sig. (2-tailed)
Y intercept	11	-1.256	0.235
Slope	11	0.309	0.317
Correlation (r)	11	1.048	0.763

Paired samples correlation

Pair	N	Correlation	Significance
Y intercept	12	0.514	0.087
Slope	12	0.157	0.167
Correlation (r)	12	0.426	0.625

Appendix 5.7 Accumulated oxygen deficit and supramaximal test parameters

Factorial Analysis

Condition = Continuous and discrete supramaximal protocol

Variable **AOD (∑L)**
By Variable **Condition**

Source	D.F.	Sum of Squares	Mean Squares	F Ratio	F. Prob.
Between Groups	1	0.288	0.288	1.584	0.224
Within Groups	18	3.272	0.182		
Total	19	3.560			

Variable AOD (∑mL.kg-1)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F Prob.
Between Groups	1	462.722	462.722	2.257	.150
Within Groups	18	3689.630	204.979		
Total	19	4152.352			

Variable Supramaximal heart rate
By Variable Condition

Source	D.F.	Sum of Squares	Mean Square	F. Ratio	F. Prob.
Between Groups	1	.800	.800	.020	.889
Within Groups	18	725.000	40.278		
Total	19	725.800			

Variable Supramaximal test time
By Variable Condition

Source	D.F.	Sun of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	995.319	995.319	.516	.482
Within Groups	18	34737.833	1929.880		
Total	19	35733.153			

Variable Supramaximal RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	7.220E-03	7.220E-03	1.109	.306
Within Groups	18	.117	6.509E-03		
Total	19	.124			

Appendix 6 Statistics Study Three

Appendix 6.1 Maximal effort comparisons for running and cycling

Factorial Analysis

Condition = exercise mode - treadmill and cycle ergometer

Variable Peak Oxygen Uptake ($L.min^{-1}$)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F Prob.
Between Groups	1	1.026	1.026	3.026	0.099
Within Groups	18	6.102	0.336		
Total	19	7.128			

Variable Peak Oxygen Uptake ($mL.kg^{-1}.min^{-1}$)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Square	F. Ratio	F. Prob.
Between Groups	1	652.310	652.310	16.182	0.001
Within Groups	18	725.608	40.312		
Total	19	1377.919			

Variable Maximal Heart Rate
By Variable Condition

Source	D.F.	Sun of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	470.450	470.450	5.784	0.027
Within Groups	18	1464.100	81.339		
Total	19	1934.550			

Variable Maximal RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	0.001	0.001	8.956	0.008
Within Groups	18	0.003	0.001		
Total	19	0.004			

Appendix 6.2 Mean percent lowest and highest submaximal exercise intensities

Factorial Analysis

Condition = exercise mode - treadmill and cycle ergometer

Variable Percent Lowest Peak Oxygen Uptake (mL.kg-1.min^{-1})
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F Prob.
Between Groups	1	245.000	245.000	2.993	0.101
Within Groups	18	1473.642	81.869		
Total	19	1718.642			

Variable Percent Highest Peak Oxygen Uptake (mL.kg^{-1}.min^{-1})
By Variable Condition

Source	D.F.	Sum of Squares	Mean Square	F. Ratio	F. Prob.
Between Groups	1	16.562	16.562	0.259	0.617
Within Groups	18	1149.760	63.876		
Total	19	1166.322			

Appendix 6.3 Predicted oxygen demand and actual oxygen uptake.

Factorial Analysis

Condition = exercise mode - treadmill and cycle ergometer

Variable Predicted oxygen demand
By Variable Condition

Source	D.F.	Sum of Squares	Mean Square	F. Ratio	F. Prob.
Between Groups	1	70.350	70.350	9.581	0.006
Within Groups	18	132.163	7.342		
Total	19	202.513			

Variable Actual oxygen uptake
By Variable Condition

Source	D.F.	Sun of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	36.342	36.342	8.750	0.008
Within Groups	18	74.763	4.154		
Total	19	111.105			

Appendix 6.4 Accumulated oxygen deficit and supramaximal test parameters

Factorial Analysis

Condition = exercise mode - treadmill and cycle ergometer

Variable AOD (∑L)
By Variable Condition

Source	D.F.	Sum of Squares	Mean Square	F. Ratio	F. Prob.
Between Groups	1	5.576	5.576	7.597	0.013
Within Groups	18	13.210	0.734		
Total	19	18.786			

Variable AOD (∑mL.kg-1)
By Variable Condition

Source	D.F.	Sun of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	2146.385	2146.385	12.965	0.002
Within Groups	18	2979.963	165.553		
Total	19	5126.348			

Variable Supramaximal Heart Rate
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	378.450	378.450	2.524	0.130
Within Groups	18	2698.500	149.917		
Total	19	3076.950			

Variable Supramaximal Test Time
By Variable Condition

Source	D.F.	Sun of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	20962.812	20962.812	9.409	0.007
Within Groups	18	40103.039	2227.947		
Total	19	61065.852			

Variable Supramaximal RER
By Variable Condition

Source	D.F.	Sum of Squares	Mean Squares	F. Ratio	F. Prob.
Between Groups	1	0.002	0.002	3.428	0.081
Within Groups	18	0.111	0.000		
Total	19	0.132			

Appendix 6.5 Pearson correlation coefficients of accumulated oxygen deficit on the cycle ergometer and the Wingate Anaerobic Test

		Bike AOD(ΣL)	Bike AOD ($\Sigma mL.kg^{-1}$)	Bike HRMax (b.min-1)	Bike Test Time (sec)	Bike RER
Wingate Peak Power (W)	Pearson Correlation	0.846**	0.388	-0.036	0.447	0.669*
	Sig. (2-tailed)	0.002	0.267	0.921	0.195	0.034
	Sum of Squares and Cross-products	492.680	4671.660	-441.000	8657.560	62.120
	Covariance	54.742	519.073	-49.000	961.951	6.902
	N	10	10	10	10	10
Wingate Mean Power ($W.sec^{-1}$)	Pearson Correlation	0.460	-0.063	-0.187	0.171	0.352
	Sig. (2-tailed)	0.181	0.863	0.604	0.636	0.319
	Sum of Squares and Cross-products	192.607	-543.606	-1644.900	2383.815	23.452
	Covariance	21.401	-60.401	-182.767	264.868	2.606
	N	10	10	10	10	10
Wingate Peak Power ($W.kg^{-1}$)	Pearson Correlation	0.739	0.538	0.417	0.784	0.847
	Sig. (2-tailed)	0.015	0.108	0.230	0.007	0.002
	Sum of Squares and Cross-products	4.731	71.093	55.970	166.523	.863
	Covariance	.526	7.899	6.219	18.503	9.592E-02
	N	10	10	10	10	10
Wingate Max HR ($b.min^{-1}$)	Pearson Correlation	0.222	0.282	0.626	0.578	0.258
	Sig. (2-tailed)	0.537	0.430	0.053	0.080	0.472
	Sum of Squares and Cross-products	12.266	321.522	724.800	1059.240	2.266
	Covariance	1.363	35.725	80.533	117.693	.252
	N	10	10	10	10	10

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).

Appendix 6.5 Pearson correlation coefficients of accumulated oxygen deficit on the cycle ergometer and the Wingate Anaerobic Test

		Bike AOD(ΣL)	Bike AOD ($\Sigma mL.kg^{-1}$)	Bike HRMax (b.min-1)	Bike Test Time (sec)	Bike RER
Wingate Peak Power (W)	Pearson Correlation	0.846**	0.388	-0.036	0.447	0.669*
	Sig. (2-tailed)	0.002	0.267	0.921	0.195	0.034
	Sum of Squares and Cross-products	492.680	4671.660	-441.000	8657.560	62.120
	Covariance	54.742	519.073	-49.000	961.951	6.902
	N	10	10	10	10	10
Wingate Mean Power ($W.sec^{-1}$)	Pearson Correlation	0.460	-0.063	-0.187	0.171	0.352
	Sig. (2-tailed)	0.181	0.863	0.604	0.636	0.319
	Sum of Squares and Cross-products	192.607	-543.606	-1644.900	2383.815	23.452
	Covariance	21.401	-60.401	-182.767	264.868	2.606
	N	10	10	10	10	10
Wingate Peak Power ($W.kg^{-1}$)	Pearson Correlation	0.739	0.538	0.417	0.784	0.847
	Sig. (2-tailed)	0.015	0.108	0.230	0.007	0.002
	Sum of Squares and Cross-products	4.731	71.093	55.970	166.523	.863
	Covariance	.526	7.899	6.219	18.503	9.592E-02
	N	10	10	10	10	10
Wingate Max HR ($b.min^{-1}$)	Pearson Correlation	0.222	0.282	0.626	0.578	0.258
	Sig. (2-tailed)	0.537	0.430	0.053	0.080	0.472
	Sum of Squares and Cross-products	12.266	321.522	724.800	1059.240	2.266
	Covariance	1.363	35.725	80.533	117.693	.252
	N	10	10	10	10	10

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).