

Abstract

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The purpose of this study was to determine the effects of an aerobic training program on the aerobic fitness and blood lipid profiles of 24 premenarcheal (PREM) and 41 postmenarcheal (POSTM) volunteers. A second intent was to test for differences in the responsiveness of the two maturity groups to the program. Subjects in each maturity level were assigned to a control (C) or training (T) group. Training consisted of 30 min continuous cycling at 75% max heart rate, three times per week for 12 weeks. All subjects were tested before the training, at week 6 and at the end of week 12. Anthropometric measurements, ventilatory threshold (VT), $\dot{V}O_2$ max and anaerobic capacity (AC), measured as total work performed during a 30 s Wingate test, were determined for all subjects at each test period. Serum total triglycerides (TG), total cholesterol (TC), low density (LDL-C), very low density (VLDL-C), and high density (HDL-C) lipoprotein cholesterol as well as subfractions HDL₂ and HDL₃ were measured pre and post training.

Analysis of variance with repeated measures revealed that both PREM groups increased their $\dot{V}O_2$ max ($p < .001$), however the increase in PREM-T exceeded that of PREM-C ($p < .01$). A training effect for $\dot{V}O_2$ max was also observed in the POSTM-T compared to POSTM-C subjects ($p < .001$) and this increase was similar to that of PREM-T. No changes in serum TG, TC, LDL, VLDL, or HDL were reported for any group. HDL₂ values decreased in all groups ($p < .001$), with

larger change occurring in the PREM subjects ($p < .001$). Although an increase in HDL₃ was observed for all groups ($p < .001$), the increase in PREM was greater than in POSTM ($p < .01$). No training effect was found in either HDL subfraction.

It was concluded that $\dot{V}O_2$ max was equally sensitive to the endurance training in both PREM and POSTM subjects. It was also suggested that, in young females, VT and AC may not be as responsive to endurance training as $\dot{V}O_2$ max. The lack of a training effect on the blood lipids and lipoproteins may be attributed to the normal concentrations in the subjects prior to the study. It is also possible that 12 weeks were insufficient to produce changes in the blood lipid profiles of the subjects.

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Dedication

To Rob,

Mon ami, mon amour, mon épouse

- and -

In memory of my Uncle Jean Labonté
who shared with me his great love of learning.

“... the search for the ultimate theory of the universe seems difficult to justify on practical grounds ... Humanity’s deepest desire for knowledge is justification enough for our continuing quest. And our goal is nothing less than a complete description of the universe we live in.”

Stephen W. Hawking

A Brief History of Time, 1988

Chapter 1

INTRODUCTION

Increased awareness of the importance of attainment and maintenance of fitness by children stems from the concept that lifetime fitness begins with exercise and health behaviors developed during childhood. Fitness has been described as the natural consequence of the important process of regular exercise (Fox and Biddle, 1988). The potential benefits of regular aerobic exercise programs in children include the optimization of health-related fitness, an increase in the quality of life, and the prevention of future disease (Pate and Blair, 1978; Shephard, 1987). Children with the experience and knowledge to maintain regular exercise as an integral part of life will be well prepared to make quality lifestyle decisions as adults (Fox and Biddle, 1988). For this reason, an improved understanding of how children respond physiologically to exercise training is essential in order to provide optimal exercise programs promoting these health benefits.

The relevance of exercise to pediatrics has begun to gain attention in the exercise physiology literature (Shephard, 1987). Pediatric investigations involving exercise now go beyond the study of the young athlete. In fact, the American College of Sports Medicine (ACSM) recently published a position statement supporting the development of physical fitness programs for children and the encouragement of healthy lifelong exercise behaviors (ACSM, 1988; Appendix A).

The relationship between regular exercise, cardiovascular health and reduced coronary heart disease (CHD) risk factors has been well documented in the adult

literature (Dufaux et al., 1982). Epidemiological studies support the concept that regular exercise is associated with a reduced incidence of CHD (Frohlicher et al, 1980) and a decrease in a number of risk factors related to the development of CHD such as abnormal blood lipid profiles, high blood pressure and obesity (Frohlicher et al, 1980; Howley et al., 1982). Regular physical activity appears to have a protective role against the development of CHD (Powell, 1987). While diseases like CHD do not normally manifest themselves during childhood, many of these risk factors originate during the pediatric years (Kannel and Dawber, 1972). Thus, physical activity in children should be encouraged and supported.

The majority of research in the field of pediatric exercise physiology has involved young male subjects with few studies involving young females. Little is known about the responsiveness of young females to aerobic exercise. Furthermore, the role of maturation in the physiological responsiveness of young females has yet to be determined. A drop in fitness levels has been reported in adolescent females (Canada Fitness Survey, 1984) with maximal relative aerobic power gradually declining from 13 years of age (Kemper and Verschuur, 1985; Mirwald and Bailey, 1986). The reduced participation in physical activity by Canadian adolescent females (Lenskyj, 1988) may, in part, be responsible for this decrease in aerobic fitness. These observations underscore the need for an increased understanding of the responsiveness of young females to exercise. To do this effectively, it will be necessary to determine if females of different maturity status (ie. premenarcheal and postmenarcheal) respond differently to physical activity and exercise (Bar-Or, 1984).

1.1 Maximal Aerobic Power in Children

The use of aerobic exercise as a method of improving health and fitness in adults is well documented (deVries, 1986, p.290; Paffenbarger et al., 1978). As in adults, the function of the aerobic system in children has typically been assessed by measuring maximal aerobic power ($\dot{V}O_2$ max). $\dot{V}O_2$ max is an accepted objective criterion of maximal aerobic power in adults (Shephard et al., 1968). It provides a non-invasive, systemic measure of the state of the pulmonary, vascular and muscular components of the organism and offers a means of measuring maximal aerobic power.

The aerobic system of pubescent and postpubescent children has been shown to be responsive to training provided appropriate stimuli are applied (Kobayashi et al., 1978; Krahenbuhl et al., 1985). However, the ability of prepubescent children to respond to such training has been widely debated (Krahenbuhl et al., 1985). Many researchers have reported significant increases in relative $\dot{V}O_2$ max following aerobic training by children (Brown et al., 1972; Ekblom, 1969; Eriksson and Koch, 1973; Lussier and Buskirk, 1977; Rotstein et al., 1986; Vaccaro and Clarke, 1987; Mahon and Vaccaro, 1989). These data indicate that when endurance training programs are of sufficient intensity, duration, and frequency, it is possible to elicit aerobic changes similar to those seen in adults.

There have also been investigations that report no change in the maximal relative aerobic power of children following training protocols similar to the ones used in the above studies (Kobayashi et al., 1978; Daniels et al., 1978; Davies, 1980; Yoshida et al., 1980). Many of the studies that have been unable to elicit improvements in relative $\dot{V}O_2$ max in children following training have demonstrated significant increases in running performance (Kobayashi et al., 1978; Yoshida et

al., 1980; Daniels et al., 1978; Davies, 1980). These findings have led to the suggestion that $\dot{V}O_2$ max may not be as valid an indicator of training-induced alterations of maximal aerobic power in prepubescent subjects as it is in more mature individuals (Bar-Or, 1983). It is also possible that the increase in performance demonstrated in these studies was due to improved running efficiency following the training.

The lower levels of the glycolytic rate-limiting enzyme, phosphofructokinase (PFK), and lower concentrations of blood lactates reported in young males (Eriksson, 1972) have led to the concept that children have a reduced anaerobic capacity when compared to adults (Bar-Or, 1983). During exercise, as intensity approaches that of $\dot{V}O_2$ max, there is an increased reliance on anaerobic glycolytic processes for energy production. A deficiency in anaerobic mechanisms could limit the maximal aerobic power of young male and female subjects.

Bar-Or (1984) has hypothesized that improvement of an initially low anaerobic capacity could account for the enhanced performances in children even though $\dot{V}O_2$ max has not improved. Reasons for this lower anaerobic capacity have yet to be defined (Wolfe et al., 1986). It would therefore be appropriate to examine change in anaerobic capacity as a potential contributor to increases in aerobic performance in children.

1.2 Anaerobic Threshold in Children

During exercise of increasing intensities, the oxygen consumption level at which aerobic processes must be supplemented by anaerobic energy metabolism has been labelled the Anaerobic Threshold (AT) (Wasserman et al., 1973). AT, a measure of maximal aerobic capacity, has been described as a better predictor of endurance

performance in adults than $\dot{V}O_2$ max (Rhodes and McKenzie, 1984). This cardiorespiratory index is normally determined either by the abrupt increase in blood lactate concentration with increasing work intensity (lactate threshold; LT) or by the non-linear increase in minute ventilation (\dot{V}_e) as oxygen consumption continues to rise (ventilatory threshold; VT). In the few studies where AT has been measured in children, a similar relationship with endurance performance has emerged (Palgi et al., 1984; Wolfe et al., 1986). Most pediatric studies have been limited to the use of the non-invasive VT method of determining anaerobic threshold due to the ethical and methodological limitations of the invasive LT method.

In adults, VT has been reported to be a reliable and valid measure of cardiorespiratory fitness (Davis, 1985). VT has been regarded as a better predictor of endurance performance than $\dot{V}O_2$ max as it reflects the maximal capacity of the aerobic system (Davis, 1985). The study of VT (or LT) in children has primarily involved comparative studies between trained and untrained subjects (Palgi et al., 1984; Reybrouck et al., 1982; Tanaka and Shindo, 1985; Wolfe et al., 1986). Information regarding the effects of aerobic training on VT or AT in children is limited. Only two studies have examined changes in VT following aerobic training (Becker and Vaccaro, 1983; Mahon and Vaccaro, 1989). However, only Mahon and Vaccaro (1989) were able to demonstrate significant training-induced increases in VT of 10–14 year old males. The effects of aerobic training on anaerobic threshold in young females, as well as in prepubescent children of either sex, has yet to be determined. As VT (and AT) can be determined without maximal effort, it has the potential to lend much to the understanding of physiological function in children with training (Palgi et al., 1984).

1.3 Exercise and The Young Female

The majority of pediatric exercise research has involved the use of young male subjects. Relatively few exercise studies have involved young females, especially of prepubertal age. However, from the health point of view it is perhaps the young female who may benefit most from improved understanding of the influence exercise and regular physical activity may have on later life. It has been reported that up until 10 years of age young males and females demonstrate little difference in relative maximal aerobic power (Bar-Or, 1983; Shephard, 1982). However, in their longitudinal study of Saskatchewan children, Mirwald and Bailey (1986) demonstrated that the rate of growth in absolute maximal aerobic power in young females (8-10 years old) was lower than that in males of similar age even though there was no statistical difference in body weight (p.19).

In females, a gradual decline in relative $\dot{V}O_2$ max has been noted beginning at approximately 12 years of age (Astrand, 1952; Kemper and Verschuur, 1985; Shephard, 1982). An age-related trend in males is not as well defined as that in females. Relative $\dot{V}O_2$ max in young males has been reported to increase until approximately 18 years of age (Astrand, 1952), remain constant (Bar-Or, 1983; Cunningham et al., 1984; Vanden Eynde et al., 1988), or decline steadily from 13-16 years (Mirwald and Bailey, 1986; Rutenfranz et al., 1981).

The changes in relative $\dot{V}O_2$ max in adolescent females may be attributed to biological factors including the increased deposition of adipose tissue characteristic of maturing females. Environmental factors such as reduced participation in physical activity as young females reach their teen years (Canada Fitness Survey, 1984) may also contribute to the decrease in maximal relative aerobic power. Vanden Eynde et al. (1988) demonstrated that activity level plays an important role

in the maintenance of maximal aerobic power during adolescence. The decline in relative aerobic power in adolescent females is probably related to a combination of both biological and sociocultural factors.

1.4 Growth, Maturation and Trainability

Trainability is the degree of functional and morphological change in an individual undergoing a conditioning or training program (Bar-Or, 1984). Thus, a solid understanding of growth and maturational influences on the physiological variables associated with the aerobic system is essential prior to evaluating changes induced by physical training.

A major methodological dilemma in pediatric exercise physiology is how to determine and quantify the relative influences of growth and exercise on children. Unlike adults, children are in a constant structural and functional flux. The simultaneous effects of growth and maturation may actually be greater than those brought about by an exercise program (Kemper and Verschuur, 1985). For example, the changes that have been observed in $\dot{V}O_2$ max, as an index of cardiovascular fitness, during normal childhood development can be explained by growth alone (Bar-Or, 1984; Krahenbuhl et al., 1985). In an attempt to alleviate this problem many cardiovascular variables are described relative to some measure of body size. Body weight is most commonly used to correct for growth (Bailey et al., 1978; Bar-Or, 1983; Kemper and Verschuur, 1985). When considering the effects of exercise on children, the concomitant influences of growth must be considered. Difficulty arises when attempting to compare training effects on individuals at varying stages of maturity. Often, correcting for body size alone will not alleviate the effects of existing functional differences. Testing for differences in the response

to exercise of groups whose values for physiological parameters are initially different is a complex task. While statistical procedures are available to correct for initial differences between groups (ie. analysis of covariance), they are not always appropriate. In order to maintain the integrity of groups of different maturity levels, it is important that these differences be recognized and protected rather than removed.

The ambivalent results obtained from pediatric exercise studies of various age groups have led to the concept of a critical developmental age when optimal responsiveness to exercise training might be expected (Cunningham et al., 1984; Kemper and Verschuur, 1985; Kobayashi et al., 1978; Mirwald et al., 1981). These studies suggest that the adolescent growth period is the critical time for the development of maximal aerobic power. However, to date, no attempt has been made to characterize the differences in changes to aerobic parameters with training in young female subjects of different maturity levels. It has yet to be determined if prepubescent, or premenarcheal, females respond in a similar manner to endurance training as more mature, postmenarcheal females.

1.5 Physical Activity and Coronary Heart Disease

Regular physical activity and endurance type exercise have been found to be associated with elevated HDL-C and triglyceride (TG) levels (Dufaux et al., 1982). Although most studies in this field have involved adult subjects, evidence is accumulating to suggest that similar relationships between exercise and serum lipids also apply to children (Nizankowska-Blaz and Abramowicz, 1983; Zonderland et al., 1984).

Atherosclerosis leading to cardiovascular diseases, though uncommon before

the age of 40 years, has been described as having origins in infancy and childhood (Kannel and Dawber, 1972). Lipid deposits, formed primarily of cholesterol, have been found in the arteries of children by the age of 3–5 years and these deposits increase in number and size with age (Kannel and Dawber, 1972). Thus, the relationship between age and the process of atherosclerotic plaque development makes the disease a pediatric concern.

Elevated low density lipoprotein cholesterol (LDL-C) level is a major coronary risk factor and is associated with an acceleration in the development of atherosclerotic plaques in humans (Rhoads et al., 1976). In contrast, an elevated high density lipoprotein cholesterol (HDL-C) level demonstrates an inverse relationship with the development of coronary heart disease (CHD) risk (Tran et al., 1983; Work, 1987). Closer inspection of the alterations to HDL-C indicate that differential changes in the HDL-C subfractions occur with exercise. The HDL₂ fraction appears to be more affected by exercise intervention than HDL₃. HDL₂ has also been identified as a stronger CHD risk factor than HDL₃ (Gidez and Eder, 1984). Much of the recent work in the field of exercise and cholesterol now include the study of changes in these HDL subfractions as a method of more clearly understanding the means by which exercise reduces the risk of CHD.

Cross-sectional data indicate that HDL-C levels are similar in young males and females (Jaross et al., 1981; Morrison et al., 1979). The first gender differences appear around puberty when HDL-C levels decline in males (Beaglehole et al., 1980). Lipid profiles of children have high predictive value for those in later life (Moll et al., 1983). These findings underline the necessity of taking preventative action at a young age. Nizankowska-Blaz et al. (1983) have suggested that prophylactic measures in children may be of importance for preventing atherosclerosis. The speculation that regular physical activity during childhood will contribute to the

prevention of CHD in adulthood (Zonderland et al., 1984) has led to a number of questions in the field of pediatric exercise physiology. Given that adherence to regular physical activity in later life is influenced by childhood experience, it is necessary to investigate the effects of exercise on blood lipid profiles in normal healthy children. To date, the majority of pediatric studies that have considered the influence of exercise on serum lipid profiles have involved athletic populations. Comparisons between athletic and non-athletic groups make up the vast proportion of the literature. Results from the few pediatric training studies available, that have involved the measurement of blood lipid and lipoproteins, have been equivocal.

A better understanding of physical conditioning of children is important for several reasons:

1. The influence of exercise on CHD risk factors in children may result in a lower incidence of the disease manifesting itself in adulthood;
2. Effective physical education in schools requires an understanding of the means by which the cardiorespiratory fitness of children can be enhanced. Curriculum design should be compatible with the activity duration, intensity and frequency requirements of the population it services;
3. Adherence to exercise and fitness will be greater if the methods are effective and appropriate for the specific population;
4. The development of healthy habits in childhood may have a positive impact on good health practices in adulthood.

1.6 Purpose

The purposes of this study were:

1. To determine the effects of aerobic training on anthropometric, cardiorespiratory and metabolic measures in premenarcheal and postmenarcheal subjects.
2. To determine if differences in the effects of aerobic training on the anthropometric, cardiovascular and metabolic measures exist between premenarcheal and postmenarcheal subjects.
3. To determine the effects of aerobic training on blood lipids and lipoprotein cholesterol levels in premenarcheal and postmenarcheal subjects.
4. To determine if differences in the effects of aerobic training on blood lipids and lipoprotein cholesterols exist between premenarcheal and postmenarcheal subjects.

1.7 Hypotheses

The following null hypotheses were tested:

H₀-1: The 12 week training program will have no significant effect on any anthropometric variable measured in the premenarcheal or postmenarcheal subjects.

H₀-2: The 12 week training program will have no significant effect on the the following cardiorespiratory variables measured in the premenarcheal or postmenarcheal subjects:

H_o-2a: Maximal Aerobic Power ($\dot{V}O_2$ max)

H_o-2b: Ventilatory Threshold (VT)

H_o-3: The 12 week training program will have no significant effect on anaerobic capacity in the premenarcheal or postmenarcheal subjects.

H_o-4: The 12 week training program will have no significant effect on the following blood lipids and lipoprotein cholesterol of the premenarcheal or postmenarcheal subjects:

H_o-4a: Serum total triglycerides (TG) and total cholesterol (TC)

H_o-4b: Very low density lipoproteins (VLDL), and low density (LDL-C) and high density (HDL-C) lipoprotein cholesterol

H_o-4c: HDL-C subfractions of HDL₂ and HDL₃

H_o-5: No differences in the effects of the training program on the anthropometric variables will exist between the premenarcheal and postmenarcheal subjects.

H_o-6: No differences in the effects of the training program on the following cardiorespiratory variables will exist between the premenarcheal and postmenarcheal subjects:

H_o-6a: Maximal Aerobic Power ($\dot{V}O_2$ max)

H_o-6b: Ventilatory Threshold (VT)

H_o-7: No differences in the effects of the training program on anaerobic capacity will exist between the premenarcheal and postmenarcheal subjects.

H₀-8: No differences in the effects of the training program on the following blood lipids or lipoproteins will exist between the premenarcheal and postmenarcheal subjects:

H₀-8a: Serum total triglycerides (TG) and total cholesterol (TC)

H₀-8b: Very low density lipoproteins (VLDL), and low density (LDL-C) and high density (HDL-C) lipoprotein cholesterols

H₀-8c: HDL-C subfractions of HDL₂ and HDL₃

1.8 Definition of Terms

Adolescence: Period of life which is associated with accelerated growth in weight and height, the appearance of secondary sex characteristics and the ability to reproduce. It continues until physical growth is complete at which time the individual is considered an *Adult*. The chronological ages at which adolescence occurs varies between individuals. However, females typically reach this maturity state approximately 2 years before males (Marshall and Tanner, 1969; 1970).

Adolescent: An individual in the maturational state of adolescence.

Blood lipid profile: In the present study, this term describes the concentrations of all blood lipids and lipoprotein cholesterols.

Blood lipid: The major blood lipids are the triglycerides, free cholesterol, phospholipids and free fatty acids.

Blood Lipoproteins: Except for the free fatty acids, the lipids circulate in the blood as lipid-protein macromolecular complexes called lipoproteins (Srinivasan et al., 1978). The relative proportions of protein and lipid determines the hydrated density of these complexes. Usually, the lipoproteins are classified on the basis of this density. These carrier molecules function to supply peripheral tissues with fatty acids and cholesterol.

Children: A group of individuals who have not yet reached adulthood as indicated by blood estradiol levels. Children may be considered prepubertal, pubertal, or adolescent depending on the level of maturity achieved.

Puberty: The term *puberty* refers to a period of time marked by the occurrence of morphological and physiological changes associated with the physical maturation of a child into the adult state. There is great individual variation in the length of this period. The appearance of secondary sex characteristics typically indicates the beginning of this developmental stage, while the attainment of reproductive function marks the termination of puberty as the individual enters adulthood (Marshall, 1978). The first signs of puberty in young females include breast and/or pubic hair development. The timing of menarche is the most reliably determined pubertal event, however it often does not occur until the latter stages of puberty. While puberty refers to that time when sexual reproduction becomes possible, females may not ovulate for up to 6 months after menarche (Marshall, 1978; Marshall and Tanner, 1969). In males, the onset of puberty is marked by the appearance of secondary sex characteristics including facial, axillary and pubic hair, and the growth of the genitalia. The onset of puberty in females is typically 2 years before males (Marshall and Tanner, 1969; 1970). This term is often

used interchangeably with the term *Adolescence*.

Pubescent: An individual in the developmental stage of puberty.

Prepubescent: In this study, the term *prepubescent* refers to an individual who has not reached puberty as indicated by the questionnaire completed by each subject and by blood estradiol levels. Secondary sex characteristics have not yet appeared. Reproduction is not possible. In the present study, this maturity state is considered similar to the classification of 'premenarcheal' since blood estradiol levels of the premenarcheal subjects were rated as 'prepubescent'.

Premenarcheal: A female who has not reached menarche.

Postmenarcheal: A female whose menarche occurred at least six months prior to the commencement of the present study as indicated by the information from the questionnaires completed by each subject.

Chapter 2

REVIEW OF LITERATURE

The growing enthusiasm and participation of children in sport and exercise has led to an increased need to define and understand the influences growth and training have on the developing child. To date, the majority of research in the area of pediatric exercise physiology has utilized male populations. The few studies that have reported the effects of exercise training on young females have generally involved competitive, elite, athletic populations (Zonderland et al., 1984). Limited research has been conducted on non-athletic young females.

Decreases in fitness level commonly reported in adolescent females (CFS, 1984) and the reduced participation in physical activity by Canadian adolescent females (Lenskyj, 1988) increase the necessity of determining methods of improving, or at least maintaining, fitness in young females as they reach adulthood. Presently, little is known about the responsiveness of young females to aerobic exercise. Furthermore, the influence of maturation on physiological responsiveness of young females to exercise has not yet been determined.

The relationship between exercise, cardiovascular health and reduced coronary heart disease (CHD) risk factors has been well documented in the adult literature (Dufaux et al., 1982; Haskell, 1984; Paffenbarger et al., 1978; Wood et al., 1984). Epidemiological studies support the concept that regular exercise is associated with a low incidence of CHD (Frohlicher et al, 1980; Paffenbarger et al., 1984;

Milvy et al, 1977) and a reduction of certain risk factors involved in the development of CHD such as abnormal lipid profiles, high blood pressure and obesity (Frohlicher et al, 1980; Hartung, 1980; Howley et al., 1982). Regular physical activity appears to have a prophylactic role against the development of CHD. While diseases like CHD do not normally manifest themselves during childhood, many of these risk factors originate during the pediatric years (Kannel and Dawber, 1972). It would therefore seem appropriate that physical activity in children be encouraged so that, as the child enters adulthood, activity will be an integral part of life.

Shephard (1987) has outlined six reasons for advocating regular exercise programs for children: 1) Optimization of general development 2) Realization of physical potential 3) Realization of intellectual potential 4) Fostering of a healthy lifestyle 5) Improvement of current health 6) Prevention of future disease. The formation of a healthy lifestyle and the understanding of its importance during the developmental years may help to reduce the risk factors involved in diseases like atherosclerosis leading to CHD and contribute to a healthy way of living throughout adult life.

2.1 Problems associated with pediatric exercise studies

A major methodological dilemma in pediatric exercise physiology is how to determine and quantify the influences that growth and exercise have on children. Unlike adults, children are in a state of structural and functional flux. For example, the changes that have been observed in $\dot{V}O_2$ max, as an index of cardiovascular fitness, during normal childhood development can be explained by growth alone (Bar-Or, 1984; Krahenbuhl et al., 1985). Therefore, it is important to understand

the role of normal growth on fitness parameters. As children mature at different rates, controlling for differences in maturity becomes a very important concern. Comparisons of children of similar chronological age may not be appropriate if maturity levels are not similar (Bouckaert et al., 1974).

Physiologists interested in studying children and exercise are faced with many constraints, both methodological and ethical. Many techniques, regularly employed with adults, may not be applied to children due to ethical restrictions. Researchers are restricted in their use of serial blood sampling, muscle biopsies and X-rays to name but a few procedures performed liberally with older subjects. Generally, pediatric research is limited to non-invasive methods and procedures.

Other problems encountered when studying performance in children include changes in motivation from one test period to another and lack of motor control or skill needed to efficiently perform the task. A well documented example of the problem is the study on 6-7 year olds by Schmucker and Hollmann (1974) which showed an 8% improvement in $\dot{V}O_2$ max following a 10-day familiarization period. One of the greatest limitations found in the literature dealing with exercise and children is the lack of longitudinal studies. Most work in this area has been performed using cross-sectional data collection from children of various ages. Interpreting results of such studies is limited as they assume that children develop at similar ages and at constant rates. Longitudinal studies of the changes occurring during such a period of growth would be much more accurate in defining and differentiating between the effects of exercise and the influences of normal growth.

2.2 The Aerobic System

The effects of growth and maturation on the aerobic energy system have been more thoroughly studied than the anaerobic system. Part of the reason for this discrepancy in information and understanding of the different energy systems stems from the ease of measurement of aerobic compared to anaerobic variables. Available information has also been enhanced by the vested interest pediatricians and pediatric pathologists have with regards to the cardiovascular system. A large portion of the understanding of the development of the cardiovascular and respiratory systems comes from initial research dealing with children and disease (Bar-Or, 1983; Cooper et al., 1984a).

As in adults, the function of the aerobic system in children is typically assessed by measuring maximal aerobic power ($\dot{V}O_2$ max). This provides a gross measure of the state of the pulmonary, vascular and muscular components of the organism. $\dot{V}O_2$ max has been described as a valid and objective criterion method of measuring maximal aerobic power (Shephard et al., 1968).

2.2.1 Standardization of Measurements.

Measurement of many of the aerobic parameters in children is not devoid of special problems. For example, in order to accurately measure $\dot{V}O_2$ max, the participant must be able to exercise to exhaustion. In such tests, motivation always plays a decisive role and children may be more reluctant than adults to push themselves to the limit.

Criteria for the achievement of $\dot{V}O_2$ max in adults include a leveling or plateau in $\dot{V}O_2$ and HR despite further increases in workload, blood lactate levels in excess of 8mM (Astrand and Rodahl, 1986: p.301), and a respiratory exchange ratio

exceeding 1.15 (Issekutz et al., 1962). There are some inherent problems in the application of these criteria to children (Rowland, 1985). For example, the smaller glycolytic potential of children will lower the respiratory exchange ratio at $\dot{V}O_2$ max in such a way as to make the adult criteria inappropriate for pre-adolescent children. In addition, the lower lactate levels generally observed in children could reflect an inability to reach the criteria of 8mM. Astrand and Rodahl (1986) noted that many children failed to exhibit a plateau in $\dot{V}O_2$ even though their ventilatory volumes, heart rates and blood lactate levels were found similar to those who did achieve a plateau. While no plateau may be exhibited, it would appear that true exhaustion occurred. Cumming and Friesen (1967) suggested that a plateau may not even exist in many children. For this reason, it has often been necessary for investigators to report 'peak' $\dot{V}O_2$ rather than $\dot{V}O_2$ 'max'. In these situations, peak has been assumed to be equal to maximal oxygen consumption. However, Cunningham (1977) demonstrated that, in repeated treadmill runs to exhaustion, there was very low reliability ($r=.27$) between test results in 10 year old males who failed to achieve a plateau in $\dot{V}O_2$ on either one of the tests. The relationship between test results was significantly greater for subjects who demonstrated a plateau on both tests ($r=.74$). These findings suggest that, in many of the subjects who failed to achieve a plateau, $\dot{V}O_2$ max was not actually measured (Krahenbuhl et al., 1985).

There is much controversy concerning the optimum method of standardizing physiological data in young children. Generally, most researchers use the traditional indices of body mass and age to describe their findings. Other attempts at standardization have focused on such parameters as skeletal age, size of organs (heart, testicular volume) and lean body mass.

When discussing physical performance in relation to age, it is necessary to

realize that chronological age is a poor reference scale. For example, Bouckaert et al. (1974) found skeletal ages of 11 year old males to range from 10 to 14 years. To compare all these males based on chronological age alone would therefore be inappropriate. Within a group of children of similar age there may be a difference of 4 or more years in terms of biological age and maturity level.

The use of skeletal age as a method of standardizing physiological data is limited. The use of X-rays for the determination and description of skeletal age is contraindicated by many pediatricians and physiologists. For this reason, Shephard et al. (1980) recommended the use of lean body mass for most physiological variables. Unfortunately, the determination of lean body mass is based on many erroneous assumptions regarding body proportions and density and may cause spurious results when applied to physiological data.

The use of peak height velocity (PHV) has been proposed as the optimal method with which to express physiological factors (Cunningham et al., 1984, Rutenfranz et al., 1984). PHV is generally observed 6-18 months prior to the peak weight velocity (PWV). The delay in PWV at puberty means that the use of weight as a descriptor of maturity becomes somewhat suspect during the years around puberty. It should be noted that the use of PHV is limited to longitudinal studies where height can be monitored over a period of time in each child studied.

There are problems in determining if changes seen in children are a result of training, growth or both. Many attempts have been made to relate the development of aerobic function to structural indices of growth in order to correct for differences in metabolic size. While traditionally aerobic power in adults is expressed in relation to body mass, the changing dimensions and proportions of children have led some investigators to question the use of such practice in pediatric populations (Bailey et al., 1978; Bar-Or, 1984; Krahenbuhl et al., 1985).

In non-human adult mammals, the relationship between O_2 consumption and body mass has been described by the dimension $M^{3/4}$ (Brody, 1945). However in humans, there is speculation that such a general equation may not adequately describe this relationship between metabolic size and function (Bailey et al., 1978). Several investigators (Bailey et al., 1978; Krahenbuhl et al., 1985; McMiken, 1976) have discussed theoretical considerations for standardization of physiological data based on physical dimensions. In most cases, length (L) has been used as the measure to which other measurements are compared. Based on geometric principles, body areas and volumes should be proportional to L^2 and L^3 , respectively. $\dot{V}O_2$ max, being a measure of volume per unit time, should theoretically be proportional to L^2 where time has the dimension of L (L^3/L).

This reasoning has also been extended to the use of body mass as a means of controlling for growth-related changes in aerobic power. Assuming that mass (M) is proportional to L^3 , then L^2 is proportional to $M^{2/3}$. Consequently, since $\dot{V}O_2$ max is proportional to L^2 , then it is also proportional to $M^{2/3}$. This model is based on the assumption that body shape and composition remain constant throughout growth and maturation while functional systematic capacities are dynamic.

McMahon (1973) proposed that it was important to incorporate the effects of elastic components of biological material on proportions and metabolic rate. Bailey et al. (1978) demonstrated that by including this elastic component, the most stable size-dissociated value for $\dot{V}O_2$ max over time in young males resulted from using the height (or length) equivalent for $M^{3/4}$, that is $L^{2.25}$. The use of height rather than body mass, as the dimension by which maximal aerobic power is expressed, has the advantage of generally being independent of environmental factors such as nutrition and physical activity (Bailey et al., 1978; Mirwald and Bailey, 1986).

The use of size-dissociated values when comparing aerobic power changes in individuals over time is optimized in longitudinal studies or in cross-sectional research involving the comparison of children of different maturity levels. Presently, the use of this method in the pediatric exercise physiology literature is very limited as it has not been demonstrated to have any practical advantage over the use of body mass or lean body mass for growth-related comparisons (Bar-Or, 1983, p.6).

2.2.2 Developmental Changes in Cardiovascular Variables.

Ultrastructural and biochemical investigations have demonstrated that aerobic factors of skeletal muscle in prepubescent children differ minimally from that of normal adults. Mitochondrial densities, intracellular lipids, and succinate dehydrogenase (SDH) activity all appear to be similar in both prepubescent and untrained adult tissue (Bell et al., 1980; Eriksson et al., 1974; Gollnick et al., 1973). Relative $\dot{V}O_2$ at maximum exercise intensities or at given submaximal heart rates does increase with age to adolescence. This increase had been attributed to both central as well as peripheral changes (Cooper et al., 1984a; Cunningham et al., 1984). Attempts to estimate the relative contribution of cardiovascular functions to an improved O_2 uptake have led to the suggestion that this rise is accomplished primarily by an improved cardiac output and, to a lesser degree, by changes in the difference between arterial and venous O_2 content ($a-\bar{v}O_2$) (Bar-Or et al., 1971; Cunningham et al., 1984). It has been established that a linear increase in O_2 -pulse ($\dot{V}O_2/HR$) with age exists for both sexes when O_2 -pulse is expressed relative to body weight (Anderson et al., 1974; Krahenbuhl et al., 1985). The lowering of heart rate (HR) for a specific exercise intensity, concomitant with a

rise in $\dot{V}O_2$ seen as children get older, is made possible by an enhanced stroke volume (SV). This is supported by the existence of an inverse relationship between HR and SV in growing children and points to the importance of SV on $\dot{V}O_2$ with increasing age (Cooper et al., 1984a). An increase in O_2 -pulse has been observed up to the age of 12 and 14 years in females and males, respectively (Anderson and Ghesquiere, 1972).

Cunningham et al. (1984) described the development of functional parameters of the cardiovascular system in relation to stages of physical maturation in a longitudinal study of circumpubertal males. By describing stroke volume and $a-\bar{v}O_2$ differences in terms of peak height velocity (PHV), these authors were able to quantify the relative effects of each on changes in $\dot{V}O_2$ with maturation.

In this study, $\dot{V}O_2$ at a HR of 155 beats per minute consistently increased across the ages of -3 to +2 yrs for PHV. A trend for a slightly more rapid increase of $\dot{V}O_2$ in the year preceding PHV was suggested, although no statistical evidence was provided. Analysis of the variability occurring in $\dot{V}O_2$ revealed a greater influence of SV on the yearly gain in $\dot{V}O_2$ than of a widened $a-\bar{v}O_2$ difference. While SV mirrored the increases in $\dot{V}O_2$ at all ages, a lag in this parameter was noticed during the period of most rapid growth, -1 yr to PHV. As there is a decrease in heart size to body mass ratio in 10-18 year olds (Blimkie et al., 1980; Bouchard et al., 1977), the apparent lag in SV may simply reflect a change in this ratio. In view of the fact that the ratio of heart size to body mass continues to decline until the late teen years, the peak velocity in SV observed for 2 years post PHV may be related to enhanced left ventricular filling (Macek, 1986), reduced peripheral resistance, or improved myocardial contractility. Indeed, all of these may mediate the changes in SV.

Unlike SV, the $a-\bar{v}O_2$ difference in the males studied by Cunningham et al. (1984) increased quite dramatically during the year before PHV. Many maturity-dependent variables may be used to account for this occurrence. Increases in muscle mass associated with peak growth with concomitant changes in O_2 carrying capacity (increase hemoglobin and myoglobin) and improved skeletal muscle blood flow all would result in a greater O_2 extraction. It was concluded that asynchronous developments of SV and $a-\bar{v}O_2$ difference at various stages of growth were responsible for the age-dependent increases in $\dot{V}O_2$. Similar changes in maturing female subjects can only be speculated at this time.

Although body size and muscle mass increase considerably during growth in children, the aerobic efficiency in children appears to be regulated so that delivery of oxygen to the muscles is maintained at optimal levels. In a study of males and females (6-17 yrs) Cooper et al. (1984a) demonstrated that a systematic relationship existed between body mass and $\dot{V}O_2$ max and AT, while work efficiency was independent of body mass. This implies that the cellular mechanism of energy utilization is quite mature even in early childhood. Sprynarova (1987), following an eight year study of males, demonstrated that peak growth velocity of functional capacity ($\dot{V}O_2$ max; max O_2 -pulse) occurred during puberty at approximately the same time as that for somatic dimensions.

2.2.3 Anaerobic Threshold

In recent years, the anaerobic threshold (AT) has emerged as a measure of maximal aerobic capacity among adults (Farrel et al., 1979; Brooks, 1985; MacDougall, 1977; Sjodin, 1982). This concept was introduced by Wasserman et al.,

(1973) as a method of defining the point when metabolic acidosis and the associated alterations in gas exchange occur during graded exercise. Therefore, AT indicates the point at which oxygen supply to the working muscles is no longer adequate in meeting their energy requirements. Considerable controversy exists as to how best to measure this aerobic variable. Some methods used include inflection of minute ventilation (\dot{V}_E) called ventilatory threshold (VT) and onset of blood lactate accumulation (lactate threshold, LT). While there are some inherent problems associated with AT it does provide important information about the aerobic capacity of an individual (Davis, 1985).

Due to methodological limitations, most of the research of AT in children has involved the measurement of VT. However, the physiological mechanisms responsible for VT are not completely understood. It has been proposed (Wasserman et al., 1973) that the increased accumulation of lactic acid in plasma during exercise results in a rise in the production of CO_2 (as a result of buffering the H^+) which in turn provides a stimulus for the disproportionate increase in \dot{V}_E with regards to $\dot{V}\text{O}_2$. Based on this information, the term VT is often used synonymously with AT. This use of VT as a non-invasive means of determining the onset of metabolic acidosis has been supported by observations of the breakpoint in \dot{V}_E appearing at the same time as the lactic acid breakpoint (Davis et al., 1976). However, the assumption that the exercise-induced metabolic acidosis and VT are cause and effect has been challenged (Brooks, 1985; Gaesser and Poole, 1986; Neary et al., 1985). Neary et al. (1985) suggested that lactic acid accumulation was not responsible for the breakpoint in ventilation (VT) during progressive exercise by adult male cyclists and that any similarity in the timing of VT and LT was only coincidental. Nevertheless, high correlations between VT and endurance performance (McLelland and Skinner, 1985; Reybrouck et al., 1983) have led to the

acceptance of VT as an objective and valid index of the capacity of the aerobic system (Davis, 1985).

The anaerobic threshold concept has been recently included in studies of children and exercise (Becker and Vaccaro 1983; Cooper et al., 1984a; 1984b; Mahon and Vaccaro, 1989; Palgi et al., 1984; Paterson et al., 1987; Rotstein et al., 1986; Rowland and Green, 1989; Vande Eynde et al., 1984). AT has been described as an objective measure that is useful as an index of aerobic capacity in pediatric populations (Vande Eynde et al., 1984). Reybrouck et al. (1982) reported a strong relationship ($r=0.93$) between VT and PWC_{170} in a group of kindergarten children. This finding has been supported by many others including Wolfe et al. (1986) who demonstrated that VT was highly predictive of aerobic running performance in prepubertal females.

As discussed earlier, children have a low capacity for anaerobic energy yield, and therefore rely on their aerobic system to perform physical work. Consequently, there has been speculation that children would reach AT at a point much closer to $\dot{V}O_2$ max (Vande Eynde et al., 1984). Since the capacity for anaerobiosis increases as children get older it would seem likely that an inverse relationship between age and AT exists.

Using the disproportionate increase in $\dot{V}e$ as their criteria for AT, Cooper et al. (1984a) demonstrated a strong correlation between AT and body weight in 6-17 year old subjects. This relationship could be interpreted to imply that muscle mass is the major determinant of AT during growth. This would not be surprising as lactic acid production is closely associated with muscle mass. As expected, the highest AT values (as a percent of $\dot{V}O_2$ max) were observed in the youngest subjects. These findings concur with those of Tanaka et al. (1985), who found the lowest maximum blood lactates and highest lactate thresholds in

their youngest males (6-8 years old). Similarly, others have reported significant decreases in VT in male and female subjects (5-18 yrs) with age (Reybrouck et al., 1985; Vandu Eynde et al., 1984; Weymans et al., 1985). In prepubescent subjects, AT has been demonstrated to be highly correlated to $\dot{V}O_2$ max with correlation coefficients ranging from 0.87 (Palgi et al., 1984) to 0.92 (Cooper et al., 1984a). For this reason, it has been postulated that a submaximal measure such as AT (as either VT or LT) could be used to provide as much information about aerobic fitness as $\dot{V}O_2$ max (Palgi et al., 1984; Mahon and Vaccaro, 1989). Since it does not require a maximal effort AT, may be a more suitable criterion of cardiovascular fitness in children. It is clearly a physiological measure that is much less dependent on the motivation and willingness of a subject to give an all-out effort.

2.2.4 Aerobic Response to Training.

Many authors have questioned if the aerobic system of prepubescent subjects is responsive to training programs. 'Trainability' is the degree of functional and morphological change in an individual who undergoes some sort of conditioning or training program (Bar-Or, 1984). An understanding of growth and maturational influences on the physiological variables associated with the aerobic system is essential prior to evaluating changes induced by physical training. The simultaneous effects of growth and maturation may actually be greater than those brought about by an exercise program.

The aerobic power of pubescents and postpubescents has been shown to be responsive to exercise training. However, much controversy exists with regard to the trainability of prepubescents. While some investigators have demonstrated

increases in maximal aerobic power in prepubescent males and females following training (Brown et al., 1972; Docherty et al., 1987; Ekblom, 1969; Lussier and Buskirk, 1977; Mahon and Vaccaro, 1989; Massicotte and MacNab, 1974; Rotstein et al., 1986; Vaccaro and Clarke, 1978), others have reported little or no change in $\dot{V}O_2$ max (Bar-Or, 1984; Bar-Or and Zwiren, 1973; Daniels and Oldridge, 1971; Daniels et al., 1978; Gatch and Byrd, 1979; Gilliam and Freedson, 1980; Kobayashi et al., 1978; Schmucker and Hollman, 1974; Stewart and Gutin, 1976; Yoshida et al., 1980).

There is evidence that endurance exercise does have an effect on certain aerobic parameters in children. Gatch and Byrd (1979) found that 8 weeks of interval cycle training at 80–90% estimated $\dot{V}O_2$ max produced significant increases in both SV and O_2 -pulse in 9 and 10 year olds. This increase in O_2 -pulse following training was attributed primarily to the observed bradycardia as opposed to changes in $\dot{V}O_2$ max. Echocardiographic findings of trained prepubescent swimmers revealed a positive influence of conditioning on left ventricular interior diameter during both systole and diastole (Medved et al., 1986). Similar findings were reported bystrand et al. (1963) who attributed elevations in maximal aerobic power in female swimmers (12–16 years) to increased heart volumes.

Unlike adults, children show no marked increase in maximal $a-\bar{v}O_2$ difference following training. Eriksson (1972) hypothesized that a large SV, attained through early training, combined with the adult response of $a-\bar{v}O_2$ to training, would enhance the adult aerobic energy supply system. However, there is no empirical data to support this theory.

Eriksson et al. (1973) used a 16 week mixed intensity training program to elicit a 14% increase in relative $\dot{V}O_2$ max of young males. Following a later training study, Eriksson and Saltin (1974) demonstrated a 30% rise in SDH activity as well

as an 8% increase in $\dot{V}O_2$ max for 11 year old males. These increases are quite similar to what would be expected in untrained adults.

In an early study of top Swedish female swimmers, Astrand et al. (1963) demonstrated that absolute $\dot{V}O_2$ was approximately 10% higher than in "normal" non-competitive females of similar age and size. A significant correlation between the volume of training these subjects performed and their functional capacity, measured as $\dot{V}O_2$ max, was also reported. These findings were interpreted as indication that young females, 12-16 years of age, could respond favorably to aerobic exercise training provided that the intensity and duration were adequate.

There have been many studies that have been able to elicit improved performance in children following training without improving $\dot{V}O_2$ max (Daniels et al., 1978; Davies, 1980; Kobayashi et al., 1978; Stewart and Gutin, 1976; Yoshida et al., 1980). In some of these investigations, absolute $\dot{V}O_2$ max appeared to increase, but once adjusted for body weight these changes no longer existed (Daniels et al., 1978; Ekblom, 1969). Bar-Or (1984) hypothesized that an improved anaerobic capacity with training may allow children to work closer to their $\dot{V}O_2$ max. This could account for the enhanced performances even though $\dot{V}O_2$ max has not improved. Another explanation for such observations is an increased mechanical efficiency in the trained subject without a concomitant increase in aerobic power.

Several authors (Krahenbuhl et al., 1985; Mahon and Vaccaro, 1987; Stewart and Gutin, 1976; Yoshida et al., 1980) have suggested that prepubescent children have such high levels of physical activity that there should be relatively small variation in their maximal aerobic power. If this is true, any training program would have to require much more activity than prepubescent individuals might normally get.

Extensive research in adult aerobic training programs has led to understanding the importance of exercise type, intensity level, duration, and frequency necessary to produce increases in aerobic power (ACSM, 1978). Rowland (1985) suggested that similar adult-related criteria for at least intensity and duration need to be applied to pediatric exercise studies in order to elicit physiological training effects. Many of the results of studies examining the trainability of the aerobic system in pediatric populations are conflicting due to the failure to properly monitor and report the duration, frequency, and intensity of exercise used. Maximal aerobic power of males and females 8–16 years of age have been shown to significantly increase following regular intensive training (Astrand et al., 1963; Docherty et al., 1987; Eriksson et al., 1974; Rotstein et al., 1986; Sprynarova, 1987; Vaccaro and Clarke, 1978). These data seem to indicate that when endurance training programs are of sufficient intensity, duration and frequency, it is possible to elicit aerobic changes similar to those seen in adults. On the other hand, there have also been investigations that report no change in $\dot{V}O_2$ max of children, especially under the age of 10 years, following training protocols similar to the ones used in the above studies. (Daniels and Oldridge, 1971; Gilliam and Freedson, 1980; Kobayashi et al., 1978). Bar-Or (1984) concluded that, based on these conflicting observations, the intensity of exercise effective in producing aerobic improvements in adults may simply not be sufficient to produce such changes during the first decade of life. It has not yet been determined whether or not this is due to an inherent unresponsiveness of the young aerobic system or simply caused by the already high activity level of prepubescent subjects. In adults, the magnitude of increase in aerobic parameters as a result of training is inversely related to the initial aerobic fitness levels. A similar relationship has been described in young males (Becker and Vaccaro, 1983).

These ambivalent results have led to the concept of a critical developmental age when trainability might be expected. Kobayashi et al. (1978) noted that the greatest response to training occurred during the adolescent growth spurt. At ages greater than one year prior to PHV, little effect of training was demonstrated in young males. However, at one year prior to PHV and thereafter, an increase in relative $\dot{V}O_2$ max was noted. Mirwald et al. (1981), using data from the Saskatchewan Growth Study, observed the mean peak $\dot{V}O_2$ max velocity just following mean PHV. The main conclusion from both these studies, as well as others (Gilliam and Freedson, 1980), was that a critical period for increasing maximal aerobic power existed in adolescence, particularly during the growth spurt.

Such ambiguity in the literature has raised concern about the validity of $\dot{V}O_2$ max as an indicator of changes in aerobic fitness of preadolescents following training programs (Bar-Or, 1984; Krahenbuhl et al., 1985; Rotstein et al., 1986). The strong relationship between AT and endurance performance in prepubescent subjects has resulted in an increased use of this parameter as a means of assessing training-induced physiological changes.

However, research involving training effects on VT in children have been limited to a few studies. Many of these studies have involved the comparison of trained subjects with untrained subjects (Atomi et al., 1986; Palgi et al., 1984; Wolfe et al., 1986). While not a training study per se, Atomi et al. (1986) compared (LT) of trained and untrained 11-13 year old males. The trained subjects had higher LT which resulted in the conclusion that the lowering of LT with age may be delayed or reduced with training. The higher LT might have been the result of the habitual exercise program of the trained group. However, other

factors including the possible preselection of children in the trained group, differences in the motivation during the testing procedure, and variation in skill level of treadmill running must be considered.

The small decrease in LT described by Rotstein et al. (1986) led to the suggestion that AT (LT) was less responsive than $\dot{V}O_2$ max to training. As $\dot{V}O_2$ max increased significantly during the training it is not surprising that little change was observed when LT was reported as a percent of $\dot{V}O_2$ max. Had they described the value in absolute or relative terms an increase may have been found.

To date, only one study has successfully produced a definite increase in VT with training of young males. Mahon and Vaccaro (1989) were successful at increasing the VT of 10–14 year old males (19%) using a running program. This was a significantly greater increase than that observed in the control subjects. Earlier, Becker and Vaccaro (1983) demonstrated a 25% rise in VT of prepubescent male subjects. However, this increase did not achieve significance when compared to the increase observed in the non-training control group. Interpretation of this study is difficult due to the large improvement observed in the controls. Presently, there have been no training studies involving young females in which AT has been assessed. It remains to be determined if this aerobic parameter will be responsive to exercise training in female children.

2.3 The Anaerobic Energy Systems

2.3.1 Influences of Growth and Maturation

It is generally accepted that children have lower anaerobic power and capacity when compared to adults (Bar-Or, 1984; Blimkie et al., 1986; Inbar and Bar-Or, 1986). In fact, Bar-Or (1984) characterised children as having a greater anaerobic

than aerobic handicap. Virtually all the information regarding both the alactic and lactic components of the anaerobic energy supply has been obtained through cross sectional studies. Ideally, a longitudinal protocol would be much more appropriate at defining growth and age related changes in this energy supply system.

Alactic Component

The two sources of energy in the alactic anaerobic system consist of ATP and creatine phosphate (CP) stores in skeletal muscle. For practical reasons, as well as ethical ones, there is very little information in the literature regarding ATP and CP stores in children. Virtually all biochemical investigations incorporating muscle biopsy techniques were performed in the early 1970's by the same group of Swedish investigators. Eriksson and Saltin (1974) measured vastus muscle ATP, CP and glycogen concentrations in 4 groups of males ranging in age from 11-16 years. ATP concentrations at rest were found to be approximately 5 mmol·kg wet weight⁻¹ across all ages tested. These values are quite comparable to those observed in adults (Karlsson, 1971). Similar observations were made for CP concentration, although there appeared to be a small but gradual increase with age (Eriksson, 1980). In addition, utilization rate of both ATP and CP during exercise mirrored the rate seen in older men (Karlsson, 1971). These findings led Eriksson and Saltin (1974), as well as others (Inbar and Bar-Or, 1986), to suggest that the alactic capacity of young males is comparable to that in adults. Thus, biochemically, children would appear not to be handicapped by the phosphagen system. However, children produce much lower peak power values than adults when measured by sprint cycle tests (such as a Wingate test) as well as the Margaria step test (Grodjinvosky et al., 1980; Sargent et al., 1984;

Tharp et al., 1984). While biochemically young subjects may not differ from older subjects, there are some other major differences influencing performance that must be considered. Once again, these involve some of the methodological problems that are evident in pediatric research. In order to generate a large amount of force quickly, a subject must be capable of innervating the proper muscle fibers. Differences in firing patterns and motor control may contribute to more efficient and effective neuromuscular control in adults compared to children. However, this theory has yet to be tested in the laboratory. Lack of skill may also be a problem as skill plays an important role in the performance of these tests. For example, a young subject will find it difficult to pedal at the frequency required by the Wingate test without losing some motor control.

Using cross sectional data from Wingate anaerobic tests, Inbar and Bar-Or (1986) attempted to describe the growth effect on the anaerobic system by summarizing the relationship of peak power (PP) and mean power (MP) to chronological age. This work demonstrated that leg PP adjusted for body weight ($PP \cdot kg^{-1}$) at 10 years was significantly lower than that achieved in adulthood. These authors also reported that PP reached maximal levels late in the second decade of life.

Lactacid Component

Muscle glycogen concentration measured in 11-16 year old males was found to be as much as 40% lower than adult levels (Eriksson and Saltin, 1974). A tendency towards higher values with increasing age was reported, with the glycogen values of the oldest males being closest to those of adults (Eriksson, 1980; Karlsson, 1971). Muscle lactate concentrations have also been demonstrated to be age-dependent.

In one study, (Eriksson et al., 1971) post exercise muscle lactate concentration was only $11 \text{ mmol} \cdot \text{kg}^{-1}$ in 13–14 yr old males while the young adults of the same study had a mean value of $17 \text{ mmol} \cdot \text{kg}^{-1}$. The reduced utilization of glycogen has been attributed to a lower concentration and activity of the rate limiting enzyme in glycolysis, phosphofructokinase (PFK) (Eriksson et al., 1971; Eriksson et al., 1973). This conforms with reports of low maximum blood lactate in children (Ilmarinen et al., 1984; Lehmann et al., 1981; Rotstein et al., 1986). The smaller glycogen stores concomitant with low PFK levels suggest that, unlike the alactic system, lactic acid anaerobic energy delivery is maturity-dependent.

Attempts have been made to explain why prepubescent subjects have a lower glycolytic capacity when compared to adults. It is possible that the relationship between testosterone and rate of lactate production demonstrated in rats (Krotkeiwshi et al., 1980) may be extended to humans. The relationship between physical maturation and the production of glycolytic enzymes in skeletal muscle of young males described by Eriksson et al. (1971) provides evidence for this concept. However, it has yet to be confirmed. Other suggestions involve the lower ratio of muscle mass to blood volume in prepubescent subjects. The relatively large blood volume in children may have a diluting effect on blood lactate. The low blood lactate levels in prepubescents may simply be an artifact caused by such a diluting effect. Additionally, Macek (1986) hypothesized that a high blood flow to the liver during exercise in children (as a consequence of a lower exercise-induced sympathetic activity) would result in a greater ability for lactate removal. The low glycolytic potential in prepubescent subjects has been supported by the reduced O_2 deficit (Eriksson et al., 1971; Macek and Vavra, 1980) and shorter half-times of oxygen increase (Cooper et al., 1984a; 1984b; Macek and Vavra, 1980; Macek et al., 1984) revealed in prepubescent males and females. These

findings suggest that the ability of a prepubescent subjects to rapidly mobilize aerobic metabolism in the early phase of exercise makes a high glycolytic capacity unnecessary. Moreover, enzymatic and morphometric analyses of skeletal muscle in young males and females have concluded that, oxidatively, the tissue of young subjects does not differ from normal adults (Bell et al., 1980; Eriksson et al., 1973; Haralambie, 1982). Therefore, the lower muscle and blood lactate in prepubescent and adolescent subjects may be due to a lower total lactate formation and a smaller anaerobic energy flux (Tanaka et al., 1985). It should be noted that at least one investigation has reported maximal lactates in prepubescent subjects that are similar to adult levels. (Cumming et al., 1980). The literature presently available on this topic appears to be equivocal.

2.3.2 Anaerobic Response to Training

Few pediatric studies have investigated the effects of anaerobic training. Generally, the research in pediatric exercise physiology has focused on the effects of exercise training on the cardiovascular parameters of the aerobic system.

A series of training studies using 11-13 yr old males demonstrated significant increases in muscle glycogen following conditioning programs (Eriksson et al., 1973; Eriksson et al., 1974). In fact, a six week training program of continuous pedalling on a cycle ergometer (3 times per week, 20 min duration, 70-85% $\dot{V}O_2$ max) resulted in an 83% increase in PFK activity (Eriksson et al., 1974). While an increase of this magnitude may not be attributed to normal growth it is important to be able to quantify and separate the influence of training from that of growth. Unfortunately, as no control group was used, it is difficult to determine if the increases in ATP, CP and glycogen concentrations were caused solely by the

training.

Eriksson et al. (1973) reported significant increases in maximal blood and muscle lactates and activity levels on PFK following training in 11-14 year old males. As lactate production is closely related to muscle mass, the rise in blood lactate as well as the increased PFK activity, may be attributed to an increase in muscle mass in the participating subjects. Nevertheless, these studies do provide evidence that the lactic acid anaerobic energy system in prepubescent males is responsive to training.

A 12 week interval training program on prepubertal male soccer players (Mosher et al., 1985) also found a 38% improvement occurred in lactic acid anaerobic performance as measured by the reduced drop off time in repeat 40 yard sprints and increased sprint treadmill running times. In addition, Grodjinovsky et al. (1980) demonstrated an improved ability to perform short supramaximal exercise following a 6 week moderately intense training program in 11-13 year old males.

It would appear that the anaerobic system of young males is capable of responding to anaerobic training. However, more information is needed to qualify and quantify the changes occurring in muscle at the cellular level. It also remains to be seen if the apparent trainability of anaerobic performance is age- and/or gender-dependent. The above studies not only considered a very small, specific age group (10-13 years), but relied exclusively on male subjects.

2.4 Atherosclerosis as a Pediatric Problem

Although factors such as a vascular spasm leading to myocardial insufficiencies often are the cause of CHD, it is atherosclerotic lesions that play the largest role in the acute genesis of CHD. The evolution of atherosclerosis follows the progression from lipid deposit through fibrous plaques to atheromatous ulceration (Dufaux et al., 1982). Early experimentally-induced lesions have been demonstrated to regress almost completely with appropriate treatment (Kannel and Dawber, 1972). However, if the lipid deposition is allowed to progress, the resulting fibrous plaques become ulcerated and calcified. These processes, as well as increased vascularization of the plaque through the ingrowth of capillaries, may be irreversible.

While symptoms of CHD are not normally manifest before 40 years of age, atherosclerosis leading to CHD has been described as having origins in infancy and early childhood (Kannel and Dawber, 1972). Autopsy studies of young American military casualties in the Korean and Vietnam wars revealed that there was evidence of coronary atherosclerotic lesion development in greater than 50 percent of these young soldiers whose mean age was 22 years (Enos et al., 1953; McNamara et al., 1971). These observations provided an understanding that atherosclerotic processes begin early in life. Holman et al. (1958), found aortic fatty streaks in many children younger than 3 yrs of age and in all children studied over this age. Lipid deposits increase in both number and size with age (Dufaux et al., 1982). Epidemiological studies report that the lipid profiles of children have a high predictive value for profiles in adulthood (Lauer et al., 1988; Moll et al., 1983). The concentration of lipoprotein cholesterol in adults, particularly the high density lipoprotein cholesterol (HDL-C), appears to be a product of factors which operate

during this period of greatest growth and development (Beaglehole et al., 1980; Miller, 1984a). The relationship between age and the process of atherosclerotic plaque development makes the disease a pediatric concern and underlines the necessity of taking preventative action at a young age.

The most effective means of reducing the incidence of CHD is through the prevention of lesion development. Epidemiological and experimental studies have shown that there is a multitude of factors which have been associated with the risk of the development of atherosclerosis. These risk factors include obesity, diet, sedentary lifestyle, elevated serum lipid cholesterol, smoking, hypertension and family history (Frohlicher et al., 1980; Kannel et al., 1979; Milvy et al., 1977; Paffenbarger et al., 1978; Stallones, 1983; Strong et al., 1973) While there is much uncertainty regarding the importance of each risk factor and their interrelationships, abnormal lipid levels appear to be the common denominator in several of the factors (Linder and Durrant, 1982).

An elevated low density lipoprotein cholesterol (LDL-C) level and a decreased level of serum high density lipoprotein cholesterol (HDL-C) are two of the major risk factors of CHD. This type of blood lipid profile has been associated with an accelerated development of atherosclerotic plaques in humans (Rhoads et al., 1976). The opposite profile, one of increased HDL-C and reduced LDL-C, has been shown to have non-atherogenic protective properties. An inverse relationship between HDL-C levels and the incidence of CHD has long been recognized (Gordon et al., 1977; Miller et al., 1977; Rhoads et al., 1976; Tran et al., 1983; Work, 1987).

2.5 Lipoprotein Metabolism

Triglycerides (TG), free cholesterol, cholesteryl esters (CE), phospholipids and free fatty acids (FFA) constitute the major lipids in human blood. With the exception of FFA which are bound to albumins, the lipids circulate in the plasma as lipid-protein macromolecular structures (Srinivasan et al., 1978). Transportation of lipids in the blood occurs by the formation of supramolecular complexes called lipoproteins. Such structures are generally spherical with a surface coat of phospholipids, unesterified cholesterol and apoproteins (apolipoproteins), and an apolar core consisting of TG and CE (Zonderland, 1985). The relative proportions of protein and lipid determines the hydrated density of these complexes. Usually, the lipoproteins are classified on the basis of this density. Chylomicrons and the very low density lipoproteins (VLDL) are the lowest density and largest lipoproteins. These molecules are responsible for the bulk of TG transport. Along with the function of supplying tissues with fatty acids, these carriers also supply cholesterol to the peripheral cells for the production of membranes and hormones. Cholesterol transport is primarily accomplished with LDL and HDL. Cellular cholesterol is acquired either by *in situ* synthesis or by the uptake of lipoprotein cholesterol. While all cells have the ability to synthesize cholesterol, it appears that the exogenous source is very important (Stein and Stein, 1987).

2.5.1 Chylomicrons and Very Low Density Lipoproteins

Chylomicrons are formed in the intestine and carry dietary TG, whereas the VLDL are synthesized in the liver and transport exogenous TG from dietary sources. The fatty acids used in the formation of hepatic TG required for the

production of VLDL are derived from both the liver and the circulation (Zonderland, 1985). Therefore, VLDL formation is relatively constant even during a fasting state. VLDL is converted into a more dense lipoprotein (LDL) following the hydrolysis of TG and degradation of the particle by extrahepatic lipoprotein lipase (LPL) at the capillary endothelial surface. During this process, unesterified cholesterol is removed and transferred to HDL (Srinivasan et al., 1978).

2.5.2 LDL Formation and Function

LDL can be formed either through the conversion of VLDL or the catabolism of chylomicrons (Nestell and Fidge, 1981). In humans, the greatest proportion of plasma cholesterol is found in the LDL fraction which promotes the flux of cholesterol into peripheral cells (Barter, 1984). Extrahepatic tissues play an important role in the removal of LDL as shown in Figure 2.1. The primary route for cholesterol delivery to the cells is through receptor-mediated endocytosis of the lipoprotein (particularly LDL). The apoproteins of LDL-C interact and bind with plasma membrane receptors. Lipid and protein contents of LDL are then internalized and degraded within the cell through endocytotic processes providing a mechanism for increasing cellular cholesterol. Approximately 60% of LDL is removed from the circulation through this pathway (Brown and Goldstein, 1984). This receptor-mediated uptake of LDL-C is inhibited by high intracellular concentrations of LDL. When this occurs, the cells begin to accumulate excessive amounts of CE by way of phagocytotic processes. The resulting 'foam cell' is generally considered a precursor to atherosclerosis (Brown and Goldstein, 1984; Nestell and Fidge, 1981).

It has been shown that atherosclerotic lesions are derived from LDL-C (Walton,

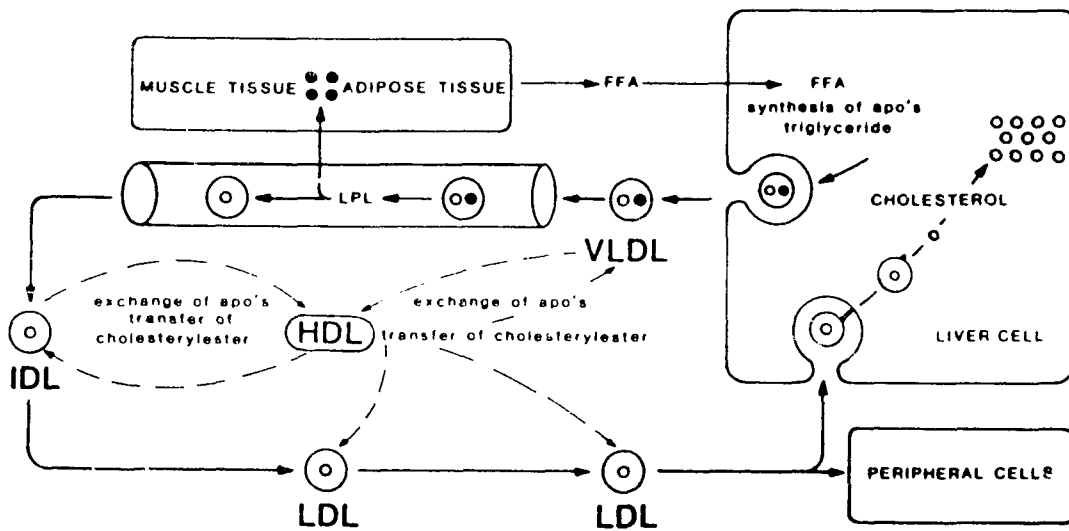


Figure 2.1: Metabolism of Low Density Lipoproteins. (From Zonderland, 1985).

1983). Additional support for this link between lesion development and concentrations of LDL-C comes from the Bogalusa epidemiological study of children, in which Neuman et al. (1986) demonstrated a significant correlation between aortic and coronary artery fatty streaks and LDL-C levels measured prior to accidental death in 35 adolescents. The formation of foam cells occurs through an uncontrolled uptake of LDL-C by macrophages and/or monocytes (Henriksen, 1984). The ability of LDL-C to infiltrate subendothelial spaces has been linked with proliferation of smooth muscle cells and the increased production of connective tissue matrices in which more lipid is deposited. The resulting injury to the endothelium, if repeated or chronic in nature, may lead to the formation of a complicated lesion. Enlargement of such a lesion could obstruct blood flow should it protrude into the arterial lumen.

2.5.3 HDL Metabolism

HDL precursors (nascent HDL) are synthesized in both the liver and intestine. HDL particles are produced within the circulation by two methods: 1) during the catabolism of VLDL when certain apoproteins are transferred to HDL and, 2) by the action of Lecithin:Cholesterol Acyl Transferase (LCAT) (Eisenberg, 1984). As depicted in Figure 2.2, HDL is a major participant in the control of lipoprotein metabolism. The primary role of this molecule is to facilitate the removal of cholesterol from peripheral tissues. HDL is the most effective lipoprotein to promote the release of cholesterol from these cells (Miller, 1984b). On the basis of *in vitro* experiments, it has been shown that HDL-C may be an anti-atherogenic agent through its competition with LDL binding and uptake to endothelial and smooth muscle cells (Stein and Stein, 1976). One of the HDL apoproteins, Apo

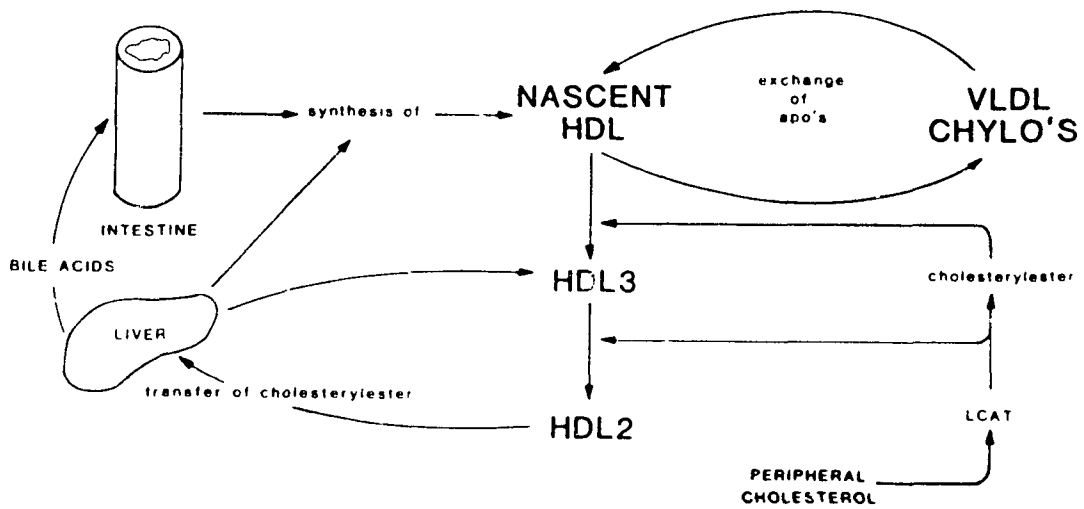


Figure 2.2: The Metabolism of High Density Lipoproteins. (From Zonderland, 1985).

A-1, acts to stimulate LCAT which, in turn, causes the esterification of cholesterol in peripheral tissues and their eventual transfer to the core of HDL (Eisenberg, 1984). Free cholesterol is continually being esterified by this plasma enzyme. Once esterified it migrates from the surface to the core of the lipoprotein. The concentration gradient formed when the lipoprotein surface becomes depleted of free cholesterol results in a net effect of free cholesterol from cell membranes moving to plasma lipoproteins, particularly the HDL (Barter, 1984). In this way HDL metabolism is believed to be central to the process of cholesterol esterification.

HDL-C Subfractions

It is the HDL-C subfraction, HDL₃, which acts as the CE recipient (Miller, 1984b). In this process, HDL₃-C is transformed to HDL₂-C and transported to the liver for subsequent catabolism and excretion as either free cholesterol or as bile acids (Reichl et al., 1982). The removal of the CE from HDL₂ allows the reconversion to HDL₃ and the continued withdrawal of cholesterol from peripheral cells (Eisenberg, 1984). HDL₃ is much more potent in promoting this net efflux of cholesterol than HDL₂ (Miller, 1984b).

In normal adults, HDL₂-C concentration is typically one third that of HDL₃-C. However, when HDL-C levels fluctuate, it is largely due to variations in HDL₂-C (Gidez and Eder, 1984). For example, the increase in HDL-C with exercise is thought to be due to a rise in HDL₂-C with no concomitant change in HDL₃-C (Wood and Haskell, 1979). In addition, cigarette smoking reduces HDL₂-C values relative to non-smokers (Elkeles et al., 1983; Stubbe et al., 1982).

Studies of individuals with coronary insufficiencies have shown that a relationship exists between low concentrations of HDL₂-C and high risk of CHD (Ballantyne et al., 1982; Miller, 1981). In support of these findings, higher levels of serum HDL₂ in fit, active male and female adults relative to sedentary individuals have been reported (Krauss et al., 1979; Kuusi et al., 1982; Laporte et al., 1983; Wood and Haskell, 1979). Although the relationship between exercise and HDL₃ remains unclear, it would appear that it is less effected by exercise than HDL₂ (Krauss et al., 1979; Wood and Haskell, 1979).

2.6 Effects of Maturation on Blood Lipids

An important influence on lipoprotein cholesterol levels in adolescents is the effects of sex hormones on the HDL molecule (Beaglehole et al., 1980). Serum HDL-C concentrations remain relatively stable until puberty (Jaross et al., 1981; Morrison et al., 1979; Srinivasan et al., 1976; Webber et al., 1983). In North America, HDL-C levels have been shown to drop 14–20% as males pass through adolescence, while in females HDL-C remains stable (Tamir et al., 1981). The result is a sharp decrease in total cholesterol (TC) in males during puberty. Following puberty, the slow increase in TG with age in males has been attributed to an increase in LDL-C rather than in HDL-C (Morrison et al., 1979). In females, TC also decreases during puberty but to a lesser extent than in males (Zonderland, 1985). While testosterone has been identified as being responsible for the decline in HDL-C in males during and post puberty, the underlying mechanisms are not well understood. In females, it has been speculated that the effects of increasing endogenous estrogens override the effects of testosterone resulting in either no change or a moderate rise in HDL-C after puberty (Beaglehole et al., 1980; Jaross

et al., 1981). However, this only occurs after females have reached menarche since the sex hormones have been reported to play no determining role on plasma lipids in premenarcheal subjects (Zonderland et al., 1986). As a results, females have consistently higher HDL-C levels than males following puberty.

Postmenarcheal, premenopausal white females have been described as being at a low risk of CHD relative to white males of similar age (Adams et al., 1987). While it is widely believed that ovarian estrogen is responsible, direct evidence for a causal relationship does not exist. Studies indicate that factors influencing endogenous reproductive steroid levels in females also influence the atherosclerotic process (Adams et al., 1987; Havekes et al., 1981). Evidence does exist for an inverse relationship between circulating levels of estradiol and extent of coronary artery atherosclerosis (Adams et al., 1987). HDL-C levels are higher in females than males after approximately 15 years of age. This has been described as being primarily due to the higher levels of HDL₂ that occur in females (Gidez and Eder, 1984). Additional evidence for the relationship between estrogen and the lipoproteins includes the increased HDL-C reported in women using oral contraceptives containing estrogens (Havekes et al., 1981).

2.7 Influence of Exercise on Blood Lipids

Several investigations have reported that the blood lipid profiles in physically active adults reflect a lower risk for CHD than blood lipid profiles of inactive reference groups (Blair et al., 1981; Durstine et al., 1987; Gidez and Eder, 1984; Haskell, 1984; Upton et al., 1984; Wood et al., 1984). Many of these studies are confounded by possible differences between those who are active (often involved in athletic competition) and those who are not. For example, there may be a

greater tendency for those who have high HDL-C levels to be involved in exercise activity. Nevertheless, the Lipid Research Clinics Prevalence Study, one of the largest data set to examine the association between exercise and the blood lipids and lipoproteins, provides evidence that in adults an increase in HDL-C with exercise occurs equally in athletes and initially sedentary individuals (Haskell et al., 1980).

Results of longitudinal training studies on sedentary individuals have been equivocal (Wood et al., 1984). While some research has been able to produce significant increases in HDL-C with exercise (Ballantyne et al., 1982; Peltonen et al., 1981), others have reported no change (Brownell et al., 1982; Dufaux et al., 1982; Lipson et al., 1980; Ready and Quinney, 1982) or even a slight decrease (Allison et al., 1981; Moore et al., 1983). However, the findings of many of these studies have been questioned due to inadequate statistical evaluation (Wood et al., 1984). Additionally, training duration may not have been sufficient in a number of studies where training lengths were as short as 6 to 10 weeks (Allison et al., 1981; Lipson et al., 1980). A dose-response relationship between activity and CHD factors including blood lipids has been proposed (Powell et al., 1987; Williams et al., 1982). It has been suggested that in adults a minimum of three months, and up to 12 months, is necessary to produce changes in lipoprotein concentrations with exercise (Pate and Blair, 1978; Wood et al., 1984). The lack of change in HDL-C with exercise may be due to the initially normal lipid profiles of the subjects at the onset of the study. It is possible that alterations would have been produced had lower initial HDL-C levels existed and the subjects been less physically active at the beginning of the studies.

As discussed earlier, cross-sectional investigations have provided some evidence that the change in HDL-C with exercise occurs through a change in the HDL₂

subfraction (Krauss et al., 1979; Kuusi et al., 1982; Laporte et al., 1983; Wood and Haskell, 1979). These findings have also been substantiated further by the longitudinal work of Ballantyne et al. (1982), who showed significantly increased HDL₂ concentration in myocardial infarction survivors following six months of exercise training. To date however, there is little clear evidence of a similar relationship with HDL₃-C values (Krauss et al., 1979; Wood and Haskell, 1979).

A number of enzyme changes may contribute to the increase in HDL-C observed with exercise. Hepatic lipase activity has been reported to correlate inversely with HDL₂-C (Kuusi et al., 1982). Some investigators have reported increases in LCAT and HDL with exercise in males (Marniemi et al., 1982) while others have failed to induce such enzymatic alterations (Thomas et al., 1985). The activity of lipoprotein lipase (LPL) in skeletal muscle, plasma, and adipose tissue has been correlated with HDL-C and in particular, HDL₂-C (Nikkila et al., 1978). However, while increases in both HDL-C and LPL activity have been demonstrated following exercise training (Peltonen et al., 1981) and in endurance trained athletes, (Nikkila et al. 1978) no significant correlation between HDL-C and LPL activity has been reported.

2.8 Effects of Exercise on Lipoproteins in Women

Most of the information regarding the effects of exercise training on serum lipids and lipoproteins has been obtained from studies on adult males. Failure of several longitudinal studies to demonstrate significant increases in HDL-C in females, despite improving aerobic performance, have led to speculation that in females HDL-C is less responsive than in males to exercise training (Allison et al., 1978; Ballantyne et al., 1981; Brownell et al., 1982; Frey et al., 1982; Lipson et

al., 1980). This diminished response has been attributed to the initially higher concentration of HDL-C in females than in males which would make changes more difficult to produce (Ballantyne et al., 1981). Presently, there are no data to support such conjecture. Another possibility for gender-related differences in response could be alterations in sex hormones. Unreported use of oral contraceptives may have obscured any training effect on the lipoproteins.

A number of cross-sectional studies comparing female athletes with sedentary females have reported significantly higher (10–25%) levels of HDL-C in the active groups compared to sedentary females (Durstine et al., 1987; Moore et al., 1983; Smith et al., 1982; Vodak et al., 1980; Wood et al., 1977). The differences in HDL-C between trained and sedentary controls are essentially the same for men and women (Wood et al., 1984). In consideration of these studies, Wood et al. (1984) proposed that it is not appropriate to associate initially high HDL-C or sex hormone levels with a potential inhibiting effect on HDL-C with exercise by women.

2.9 Exercise and Blood Lipoproteins in Children

While HDL-C levels have been reported to be higher (16–30%) in active, fit adults than in sedentary adults (Durstine et al., 1987; Moore et al., 1983; Wood et al., 1977; Hartung et al., 1980), there is no evidence that a similar relationship holds true for pediatric populations. Currently, only a limited amount of research has been directed at the influence of exercise training on blood lipid and lipoprotein concentrations in children (Bell et al., 1989; Deveaux et al., 1986; Gilliam and Burke, 1978; Gilliam and Freedson, 1980; Linder et al., 1983; Nizankowska-Blaz and Abramowicz, 1983; Parizkova et al., 1986; Savage et al., 1986; Thorland and

Gilliam, 1981; Valimaki et al., 1980; Zonderland et al., 1984). Most of these studies have not incorporated exercise training within their research design, but rather have involved the comparison of athletic populations with less active, or sedentary groups. Most of the comparative studies have demonstrated significant differences in blood lipid profiles of active and inactive males and females ranging in age from 3–17 years of age (Bell et al., 1989; DuRant et al., 1982; Nizankowska-Blaz and Abramowicz, 1983; Parizkova et al., 1986; Thorland and Gilliam, 1981; Valimaki et al., 1980; Zonderland et al., 1984). In a study of trained and untrained post-pubescent males and females, Bell et al. (1989) reported that endurance trained male and female athletes (15–17 years old) had significantly higher (8%) HDL-C levels, lower (25%) LDL-C and TG values and increased HDL-C to Total Cholesterol ratios (HDL:TC). Similar findings were reported by Zonderland et al. (1984) who compared premenarcheal gymnasts and swimmers with untrained controls of similar maturity status. As this was not a true training study, it is not possible to state conclusively that the differences between the groups were due to the training.

Results of the few exercise training studies involving blood lipid profiles of children have been equivocal. Table 2.1 provides a list of the pediatric exercise studies that have evaluated blood lipoprotein changes in children. Gilliam and Burke (1978) found significant increases in HDL-C and HDL:TC ratios in prepubescent females after six weeks of aerobic type exercise. However, interpretation is limited due to the lack of a control group. The HDL:TC ratio is used both to assess CHD risk and to observe changes induced by various intervention methods. However, it is more informative to consider each component separately. Three reported studies (Deveaux et al., 1986; Linder et al., 1979; 1983) failed to elicit alterations in any of the blood lipids or lipoproteins measured following

Table 2.1:
Aerobic Exercise Training studies involving the assessment of lipoproteins in children.

Study	Subjects	Training	Results	Comments
Gilliam and Burke (1978)	F, 8-10yrs n=14	6 weeks aerobics	↑ HDL-C ↑ HDL:TC	no controls
Linder et al., (1979)	M,F; 7-15yrs n=103 Ex (n=?) C (n=?)	4 weeks training no training	no Δ	no descript. of exercise
Linder et al., (1983)	M, 11-17yrs Ex (n=29) C (n=21)	8 weeks walk/jog summer activ.	no Δ	poor descript. of exercise intensity
Deveaux et al., (1986)	M, 14-17yrs (n=12)	8 weeks Soccer, 4/wk 1.25hrs, mod. intensity 1 game/wk	no Δ	no controls
Savage et al., (1986)	M, \bar{X} =8.5yrs High I (n=12) Low I (n=8) C (n=10)	10 weeks walk/jog/run 75% VO ₂ max 40% VO ₂ max no training	↓ HDL-C	Initial HDL-C Higher than C

various methods of training by children. These researchers questioned the low intensities and short durations (4–8 weeks) of the training programs and stated that they were probably not sufficient to produce any changes. In support of this, only one of these studies reported improvements in aerobic performance (Linder et al., 1983). However A drop in HDL-C has also been reported in prepubescent males after participation in a 10 week walk/jog/run program (Savage et al., 1986). However, the initially higher HDL-C levels in the training group in relation to the control group may have been a cause of this result.

No well controlled exercise training study employing a non-training control group has been attempted with a pediatric population. In addition, no study of the effects of exercise on the HDL subfraction HDL₂ and HDL₃ in children has been reported. While many have suggested that interventions, such as exercise, designed to modify CHD risk factors should begin at an early age (Deveaux et al., 1986; Gilliam et al., 1977; Gilliam and Freedson, 1980; Nizankowska-Blaz and Abramowicz, 1983; Zonderland et al., 1985) there is little evidence that exercise has such an effect on CHD risk factors in healthy children. It has been suggested that exercise will have no effect on blood lipid profiles in children unless their cholesterol levels are abnormal (Montoye, 1985). It remains to be determined if blood lipid and lipoproteins in children are as responsive to exercise training as they appear to be in adults.

Chapter 3

RESEARCH METHODS

As outlined in Chapter 1, the intent of this study was to assess the responsiveness of premenarcheal and postmenarcheal subjects to an aerobic training program and to determine if maturational differences exist in the responsiveness to such training. To make this possible, the following methods were employed.

3.1 Subjects

The subjects were females recruited from a local elementary school (grades 4-5) and a local junior high school (grades 8-10) in School District #61 (Greater Victoria). These two schools are within 1 km of each other and their populations are demographically representative of the community they serve. Many of the junior high school subjects had previously attended the elementary school involved in this study. A number of the families participating in the study had children attending both schools.

The purposes and procedures of the research were described to the volunteers and their parents prior to obtaining informed consent (Appendix B). The subjects were characterized as premenarcheal (PREM) or postmenarcheal (POSTM) based on information obtained through a questionnaire distributed to each subject and her parents (Appendix C) as well as through blood estradiol assessment. Those females whose menarche had occurred at least 6 months prior to the study were

included in the POSTM group. Only those subjects who had not reached menarche were included in the PREM group. All POSTM subjects were from the junior high school while all PREM subjects were from the elementary school. Mean ages were 9.8 years (SE=0.1) and 14.3 years (SE=0.1) for the PREM and POSTM groups, respectively. Subjects were also characterized as being untrained. No subject was involved in any physical training prior to the study.

Subjects in each maturity group were matched for size and performance on the laboratory pretests and assigned to either an exercise training (T) or a non-training control (C) group. The original number of volunteers totalled 27 PREM and 44 POSTM, however 5 subjects withdrew prior to the termination of the study for health or family relocation reasons: one each from PREM-C, POSTM-C, POSTM-T and two PREM-T subjects. One subject (POSTM-C) withdrew due to lack of interest. The final group samples are shown in Table 3.1.

3.2 Testing

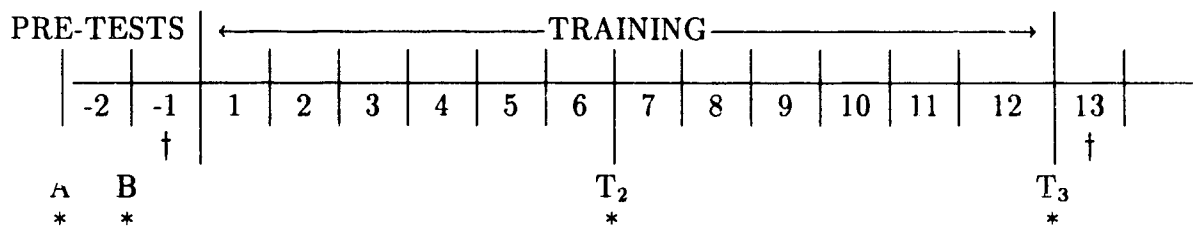
All testing was performed in the Sport and Fitness Testing Center at the University of Victoria, with the exception of the Wingate Anaerobic tests (WAnT) which were performed at the schools. During their first visit, subjects were provided with a description of all testing protocols and were familiarized with the procedures.

The testing schedule involved two pretests, a mid-study test at the end of week 6 (6wk) and a final test at the end of week 12 (12wk) as described in Figure 3.1. The two laboratory pretests were performed by all subjects as a means of familiarizing the subjects with the methods. These pretests were separated by 5-7 days. Both pretest periods involved the anthropometric assessment and the $VT/\dot{V}O_2$

Table 3.1: Subjects Divided By Maturity and Experimental Group.

Experimental Group	Maturity Group		N
	PREM	POSTM	
Control	12	20	32
Training	12	21	33
Total	24	41	65

Figure 3.1:
Schedule of Testing (weeks)



* Anthropometry and $\dot{V}T/\dot{V}O_2$ max cycle tests.

† Blood collection and Wingate Anaerobic cycle test (at the school).

max cycle test. For data analysis purposes, the highest $\dot{V}O_2$ max of the pretests and their corresponding VT were used as the PRETRAIN values. Individual anthropometric PRETRAIN values were calculated as the mean of the results from each pretest. In the week following the second pretest, the first blood collection and Wingate anaerobic cycle tests were performed at the schools on separate days and these tests were not duplicated. The training program began one week after the second set of pretests.

3.3 Laboratory Procedures

3.3.1 Anthropometry

Upon entering the laboratory each time, mass and height were measured according to the methods of the Canadian Standardized Test of Fitness (CSTF, 1987). Duplicate skinfold measures were taken at 5 sites: triceps, biceps brachii, subscapularis, iliac crest, and medial calf using Harpenden steel calipers (British Indicator Ltd., UK) as described by the CSTF (1987). Percent body fat (%BF) was determined using the method of Durnin and Womersley (1974). Lean body mass (LBM) was calculated as the actual body mass minus the mass of the fat measured by skinfolds:

$$LBM = mass(kg) - (mass * \%BF)/100$$

The estimation of %BF from anthropometry in children and youth is limited by a number of problems. The primary source of error involves the lack of cross-validation of anthropometric equations for youth populations for assessment of applicability to general pediatric populations. The equations utilized for the estimation of %BF in children are, for the most part, based on adult models. The

results obtained by such equations have been reported to yield an overestimation of fatness and, as such, an underestimation of LBM (Boileau et al., 1985). The validation of anthropometric methods in adults involves cross-validation with criterion body composition techniques including densitometry and ^{40}K spectroscopy. The invasiveness of these methods limit their application to children. Thus, due to ethical reasons, the validation of skinfolds as a means of determining %BF in children is hindered. In the present research, the intent of the anthropometric measurements was to monitor for body composition changes over the duration of the study. As the methods remained consistent over the period of the study, a comparison of results from one time period to the next is acceptable. The potential for overestimated %BF values, especially for the premenarcheal subjects, must be considered when interpreting the results.

3.3.2 Ventilatory Threshold and Maximal Aerobic Power

A cycle ergometer test involving an incremental continuous protocol was used to measure ventilatory threshold (VT) and maximal aerobic power ($\dot{V}\text{O}_2 \text{ max}$). All tests were performed on a manually braked Monark cycle ergometer equipped with toe clips to improve pedalling efficiency. Respiratory and metabolic measures were monitored every 30 s using a Beckman Metabolic Measurement Cart (MMC). These measurements included minute ventilation (\dot{V}_e), fractions of expired CO_2 and O_2 , volumes of produced CO_2 ($\dot{V}\text{CO}_2$), and O_2 ($\dot{V}\text{O}_2$); and respiratory exchange ratio (R). The MMC was calibrated with known primary standard gases before and after each test. Heart rate (HR) was telemetered every minute using a Sport Tester (PE3000) monitor.

Following a 2 min warm-up of unloaded pedalling, resistance was increased

by a minimum of 15 watts every 2 min until a non-linear increase in \dot{V}_e vs $\dot{V}O_2$ occurred in a protocol modified from James (1978). The magnitude of increase in resistance was dependent upon the body mass of each individual and ranged from .25kp to 1.0kp. From this point, increments in resistance were applied every minute. Subjects were instructed to remain seated for the duration of the test and were encouraged to continue until they could no longer maintain the pedal frequency at 60 rpm. A plateau in $\dot{V}O_2$ while resistance continued to increase (a rise of $<2 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) was the primary criteria used to indicate that $\dot{V}O_2$ max had been reached. If no plateau was achieved, subjects were required to attain the following two secondary criteria in order for 'peak' $\dot{V}O_2$ to be considered $\dot{V}O_2$ max: 1) HR >200 bpm; 2) R >1.15 . All but one subject were able to fulfill at least two of these criteria for each of the test periods. This PREM-T subject (SS) did not successfully meet the $\dot{V}O_2$ max criteria in the final test (12wk). For this reason, the data analysis for $\dot{V}O_2$ max and related variables involves $n=11$ for the PREM-T group. This subject did complete all other requirements, therefore her results were included in all other data analyses.

$\dot{V}O_2$ max was expressed in a number of ways: in absolute terms ($\text{l} \cdot \text{min}^{-1}$); relative to total body mass ($\text{ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$); and relative to lean body mass ($\text{ml} \cdot \text{kg}(\text{lbm})^{-1} \cdot \text{min}^{-1}$). Size-dissociate values for $\dot{V}O_2$ max were also determined using height^{2.25}, the height (length) equivalent for Mass^{3/4} (Bailey et al., 1978).

Application of a multi-segmental non-linear regression technique (Jones and Molitoris, 1984) was used to locate the 'breakpoint' in \dot{V}_e as $\dot{V}O_2$ was increasing. The method utilized an approximate F test to assess the fit of the produced 'broken' regression line to the data of each individual subject. A significant F would indicate that the broken line was a significantly better fit than a single

straight line. This breakpoint was interpreted as VT and described as $\dot{V}O_2$ at VT as well as a percent of $\dot{V}O_2$ max. Once VT was determined using this procedure, the results of each subject for all tests were verified visually against the raw data.

3.3.3 Anaerobic Capacity

Wingate anaerobic cycle tests (WAnT; Bar-Or, 1987) were administered to all subjects within 5 days of each aerobic test as a means of assessing anaerobic capacity. The protocol involved a 30 s supramaximal effort at a resistance setting of 0.075 kp· kg body wt⁻¹. Toe clips were used to improve pedal efficiency and subjects were instructed to remain seated. Verbal motivation to maintain maximal pedalling rate throughout the test was provided by the testers during all WAnT. A micro switch attached to a pedal on the ergometer and connected to a chart recorder was used to measure pedal frequency. Total work (joules) was calculated over the 30 s and used to reflect anaerobic capacity (AC) in absolute terms. AC was also corrected for body size and expressed relative to body mass (ACrel, joules· kg⁻¹) and lean body mass (ACl_{lbm}, joules· kg(lbm)⁻¹).

3.3.4 Blood Collection

The study involved two blood collection periods: 1) PRE test, before the T groups began training and 2) POST test, during week 13. Following a 12 hour overnight fast, blood was collected from each subject at the antecubital vein by an experienced pediatric laboratory technician. Venipuncture was performed using 22 gauge stainless steel needles while the subjects remained in a sitting position. A tourniquet was placed approximately 2 inches above the elbow but released before 14–21 ml venous blood were drawn from the antecubital vein into a set of

7ml vacutainers. Early tourniquet release was used to avoid an artifactual increase in the concentration of plasma lipids (Bachorik, 1982). Once blood collection was completed the subjects were provided with breakfast including dairy products, juice and muffins.

3.3.5 Blood Lipid Analysis

All blood analyses were performed at Island Medical Laboratories in Victoria, B.C. Blood samples were stored at -20°C for a maximum of 14 days. Prior to analysis, samples were allowed to thaw completely at room temperature. Samples to be analyzed for lipoproteins were also stored in the dark at -20°C and analysed within a few days of collection. It has been suggested that the fragile lipoproteins do not tolerate freezing for long periods without undergoing alterations in their ultracentrifugal characteristics (Bachorik, 1982). However, Gidez et al., (1982) demonstrated that storage of plasma at -20°C for short periods (7–30 days) is preferable to storage at 4°C when a dual precipitation method is to be applied. Since this was the method used in the serum analysis, it was felt to be appropriate for this study.

All analyses for lipids and lipoproteins were made using the serum of each sample. It has been determined that cholesterol does not exist inside erythrocytes and that the cholesterol found in the membranes of the red blood cells exist in equilibrium with the serum pool of free cholesterol (Cooper, 1970). Additionally, the use of serum rather than plasma can alleviate certain problems caused by the endogenous anticoagulants found in plasma (Zak, 1977).

Total Serum Triglycerides

Serum triglycerides (TG) were analyzed using a commercial kit (Diagnostic Chemicals, Ltd, Charlottetown, PEI, cat. no. 210-75) that employed the enzymatic reaction of L- α -glycerol phosphate oxidase (GPO) described by Nagele et al., (1984). This was followed by a colorimetric measurement of the resulting quinoneimine by a Cobas Bio random assess analyzer (Roche, Co.). This is the routine method for serum total triglyceride analysis at Island Medical Laboratories. All samples were assayed in duplicate with a reported coefficient of variation (CV) of 2.8%. This CV is within the 'ideal' precision range as recommended by the Laboratory Standardization Panel of the National Cholesterol Education Program (1988). Appendix D describes the details of this method.

Total Serum Cholesterol

All samples were assayed enzymatically for total cholesterol (TC) using a commercial kit (Diagnostic Chemicals, Ltd, Charlottetown, PEI, cat. no. 225-26) and a colorimetric measurement of the resulting quinoneimine dye by a Cobas Bio random assess analyzer (Roche, Co.) following the method of Allain et al.,(1974). It is the routine method of cholesterol analysis at Island Medical Laboratories. All samples were assayed in duplicate with a reported coefficient of variation (CV) of 3.2%. This CV is within the 'ideal' precision range as recommended by the Laboratory Standardization Panel of the National Cholesterol Education Program (1988).

This procedure was used to determine the cholesterol content of High Density Lipoproteins (HDL) as well as in the HDL subfractions HDL₂ and HDL₃. The details of this procedure can be found in Appendix E.

High Density Lipoprotein Cholesterol

HDL-C was determined using two different methods. The results of the first method, using phosphotungstic acid (PTA), were used to calculate the low density lipoprotein cholesterol (LDL-C) and very low density lipoprotein (VLDL) concentrations. The second method was a dual precipitation technique performed in order to separate the two HDL subfractions. With both methods, the measurement of cholesterol in each supernatant produced was made using the total serum cholesterol method described above. The HDL-C results obtained with the PTA method were the data used in statistical analysis of HDL-C.

A. Phosphotungstic Acid Method

The phosphotungstic acid method (PTA) is the routine procedure used by Island Medical Laboratories for the determination of serum HDL-C. LDL and VLDLs were precipitated from serum by the addition of a HDL-precipitating reagent containing PTA and Mg^{2+} as described by Dias et al., (1988). The precipitate containing LDL and VLDL was removed by centrifugation (2000 x g, 30 min at 4°C) leaving the HDL-C in the remaining supernatant. The total serum cholesterol (TC) assay described earlier was then applied to the supernatant in order to determine the cholesterol content in the HDL fraction of the original sample.

B. Dextran Sulfate Method

This technique for the determination of the HDL subfractions HDL₂ and HDL₃ involved two precipitation procedures utilizing a solution of dextran sulfate (DS; M_r 50,000 ± 5,000, Northwest Lipid Research Group, Seattle, WA.) and MgCl₂

(0.6 mol·l⁻¹) as the precipitating agent (Talameh et al., 1986). The first precipitation involved the fractionation of total HDL. Addition of the DS-MgCl₂ to serum resulted in the precipitation of LDL and VLDL following centrifugation (1500 x g, 15 min, 4 °C). The supernatant was then assayed for HDL-C using the TC method described above. The second precipitation involved the fractionation of HDL₂ from an aliquot of supernatant produced by the first precipitation. In this procedure a stronger concentration of MgCl₂ (3 mol·l⁻¹) was used in the DS-MgCl₂ precipitation agent resulting in the precipitation of HDL₂. Immediately following centrifugation (1500 x g, 1 hr, 4 °C) the supernatant containing HDL₃ was removed and analyzed for cholesterol using the TC method described above. HDL₂-C concentration was then calculated as the difference between the total HDL-C (DS-MgCl₂ supernatant) and the HDL₃-C.

Low Density and Very Low Density Lipoprotein Cholesterol

LDL-C and VLDL-C levels were calculated using the Friedewald equation (Friedewald et al., 1972) following the determination of HDL-C using the PTA method. This equation requires the measurement of TG, TC and HDL-C and assumes a fixed relationship between VLDL-C and TG:

$$LDL - C(mm\text{ol} \cdot l^{-1}) = TC - (TG/2.2) - (HDL - C, PTA)$$

where,

$$VLDL - C(mm\text{ol} \cdot l^{-1}) = TG/2.2$$

This method has been found to be a simple, yet reliable and convenient way of estimating LDL-C and VLDL-C provided TG values are below 3.5 mmol·l⁻¹

(Rao et al., 1988). All TG values for the subjects in this study were below this level.

Blood 17 β -Estradiol levels

Serum samples used for hormone analysis were frozen at -20°C for a maximum of 21 days prior to being assayed for 17 β -Estradiol. Prior to analysis, samples were thawed at room temperature.

Serum 17 β -Estradiol (Estra-1,2,5(10)-triene-3, 17 β -diol) was measured using a time-resolved fluoroimmunoassay kit (DELFLIA, Walla, Finland Kit no. 1244-024). This assay is a solid phase fluoroimmunoassay, based on the competition between europium-labelled estradiol and sample estradiol for polyclonal anti-estradiol antibodies derived from rabbit immunoglobulin G (IgG; Hemmila et al., 1984). A second, anti-rabbit, IgG antibody was used to coat the solid phase (walls of the mixing wells) and bind the IgG-estradiol complex, thereby providing a rapid and complete separation of antibody-bound and free antigen. The assay required one incubation step of 2 hrs at room temperature with slow shaking. The higher the concentration of estradiol in the sample, the lower the concentration of europium-labelled estradiol bound to the solid phase.

Commercial standards of known concentrations ranging from $.05\text{ nmol}\cdot\text{l}^{-1}$ to $15\text{ nmol}\cdot\text{l}^{-1}$ were used. Samples were assayed in quadruplicate. Intra assay precision with the range of the standard curve was 4.5% (CV). This is an acceptable CV for the method utilized. Results were converted from $\text{nmol}\cdot\text{l}^{-1}$ to $\text{pmol}\cdot\text{l}^{-1}$ for ease of interpretation. This method was accurate to as low as $50\text{ pmol}\cdot\text{l}^{-1}$ ($.05\text{ nmol}\cdot\text{l}^{-1}$) and therefore any sample containing less than $50\text{ pmol}\cdot\text{l}^{-1}$ was reported as $<50\text{ pmol}\cdot\text{l}^{-1}$. The following normal ranges were used to determine

the maturity classification for each sample. These ranges were developed from the database of Island Medical Laboratories reflecting values for a population similar to the one involved in this study:

Pre-Pubertal Child	less than 90 pmol·l ⁻¹
Normal Adult Female:	
Mid-Follicular Phase	110–584 pmol·l ⁻¹
Ovulatory Peak	550–1650 pmol·l ⁻¹
Mid-Luteal Phase	550–845 pmol·l ⁻¹
Pregnancy	up to 70,000 pmol·l ⁻¹

3.4 Dietary and Physical Activity Records

While subjects participating in this study were not required to adhere to a specific diet, they were asked to maintain a consistent diet throughout the study. In order to accurately determine the effects of the training protocols used in the study, the subjects in the control groups were instructed to maintain their pre-study physical activity patterns and to avoid any extra-curricular activities that might induce a training effect. The subjects in the training groups were asked that the addition of the training sessions of the study be the only change to their habitual physical activity patterns. To monitor the diets and activity levels of the subjects, each was required to complete a short questionnaire when they arrived for each testing session (Appendix C). These questionnaires indicated that no subject altered her diet or activity patterns in such a way that she was no longer eligible to participate in the study.

3.5 Training

All training occurred in designated areas at the two respective schools. The 12 week training program consisted of continuous cycling three times a week for 30 min at 75% of determined max HR. Heart rates were monitored during each training session using Sport Tester (PE3000) monitors as well as radial or carotid pulse. All T subjects were provided with water and/or fruit juice at each session. In order to encourage attendance, video taped movies were shown at most training sessions (Appendix F). Attendance at the training sessions was 89.5% and 91.3% for the PREM-T and POSTM-T groups, respectively.

3.6 Motivation.

All participants received a T-shirt identifying them as being involved with the study. In addition, prizes donated by local merchants were distributed randomly throughout the course of the study (A list of sponsors can be found in Appendix G). To ensure that the control groups returned at the end of the 12 weeks, each C group subject who performed the final tests had her name entered in a draw for a monetary prize. In order to motivate the T group subjects to continue for the last 2 weeks of training, they were told that their names would be entered in a separate draw for a monetary prize for every session they attended. Separate draws were held for each T and C group.

3.7 Statistical Analysis

The following factorial design approach was utilized to analyze the data:

1. The initial analysis involved a 2 (Maturity) \times 2 (Training level) \times 3 (Tests) factorial design analysis of variance (ANOVA) with repeated measures for each of the dependent variables. Alpha was set a priori at the .05 level for significance. This analysis was utilized to test for differences between the premenarcheal and postmenarcheal maturity groups and between the training and control groups. It was also used to determine the effects of Test and the interactions between Maturity level and Test, and Training level and Test. When a significant interaction between two of the independent variables was indicated a series of 2 \times 3 ANOVA were employed to locate where the differences existed.
2. Based on the finding of a significant Maturity level by Test interaction, subjects were divided by Training level and a 2 \times 3 (Training level by Test) ANOVA was used in order to locate the maturational differences within each training group. When a significant Training level by Test interaction was indicated in the initial analysis, the subjects were separated by Maturity level followed by a 2 \times 3 (Maturity by Test) ANOVA employed to find the differences between the training and control groups of each maturity level.

The findings of analysis (2) would identify any maturity-related responsiveness to the training program, as well as the existence of any training effect within each of the maturity groups.

When the initial analysis was used to test for differences in anaerobic capacity (AC), it was determined that the two premenarcheal groups differed significantly

throughout the study. Therefore, analysis of covariance (ANCOVA) with PRE-TRAIN scores as the covariate was used to accurately compare the means of these two groups. This analysis followed similar procedures as described above for the ANOVA.

3.7.1 Tests for Reliability of Methods

The two $\dot{V}O_2$ max pretest scores were used to produce a test-retest reliability coefficient through the application of a Pearson Product-Moment correlation procedure. This procedure was used to determine the position stability of individual scores within the subject sample population. Following this, a paired t-test was used to test for differences between the means of the two pretest results. Lack of any significant difference between the scores would reflect intersubject score consistency over time. Similar statistical procedures were used to determine the reliability of the VT and WAnT methods. Only one pretest WAnT was performed, therefore only the C group WAnT results at PRETRAIN and 6wk were used in this analysis. Results of the tests for reliability are given in Appendix H.

Chapter 4

RESULTS

Tests for homogeneity of variance (BoxsM; SPSSX) between groups were not significant ($p > .05$).

4.1 Anthropometry

No differences existed between the T and C groups within each maturity level at any of the test periods of the study. Table 4.1 summarizes the findings for all anthropometric measures.

4.1.1 Body Mass

The 2 x 2 x 3 ANOVA results demonstrated that the PREM subjects were significantly lighter ($F(1,61)= 81.80, p < .001$) than the POSTM subjects at each test period. A significant Maturity by Test interaction ($F(2,122)= 4.33, p < .05$) was found for body mass indicating that one Maturity level demonstrated greater changes in mass than the other. When subjects were separated by Maturity level, a Test main effect for both PREM groups was found ($F(1,22)= 7.88, p < .001$). The significant increase in mass demonstrated by both PREM groups indicates a growth effect rather than a training effect. This is supported by the lack of a Training level main effect for body mass within the PREM groups. No similar observation was made for the POSTM subjects as their mass remained constant over the 12 weeks of the study.

4.1.2 Height

The PREM subjects were shorter than the POSTM subjects throughout the study ($F(1,61)= 164.53, p <.001$). Similar to mass, a significant Maturity by Test interaction existed for height ($F(2,122)= 39.32, p <.001$). Subsequent analyses of the data following division of the subjects by Maturity level found that both PREM groups increased their height by a similar magnitude ($F(1,22)= 68.28, p <.001$). Height remained unchanged in the POSTM subjects (Table 4.1).

4.1.3 Sum of Skinfolds

No differences in the sum of skinfolds (SF) existed between the Maturity or Training groups throughout the study (Table 4.1). This finding was consistent for both the sum of five skinfolds described by CSTF (1987) and the method of Durnin and Wormersly (1974). The POSTM subjects did demonstrate slightly higher SF values, yet the large range of scores observed for all groups resulted in non-significant findings for this dependent variable.

4.1.4 Percent Body Fat

Percent body fat (%BF) was predicted from the SF derived from the method of Durnin and Wormersly (1974). The PREM subjects had a lower percent body fat (%BF) than the POSTM subjects as demonstrated by a significant Maturity main effect ($F(1,61)= 5.11, p <.05$). %BF scores remained constant for all groups for all test periods (Table 4.1).

Table 4.1:
**Means and SE for Anthropometric Variables Measured Over
 Three Tests for the Premenarcheal and Postmenarcheal subjects.**

		MASS (kg)						
		n	PRETRAIN		6wk		12wk	
PREMENARCHEAL ^a								
Control	12	36.0	(1.5)	36.7	(1.5)	36.5	(1.8) ^b	
Train	12	36.6	(2.5)	37.5	(2.5)	37.6	(2.4) ^b	
POSTMENARCHEAL								
Control	20	55.4	(1.5)	55.5	(1.3)	55.3	(1.4)	
Train	21	57.3	(2.4)	56.8	(2.4)	57.3	(2.3)	

		HEIGHT (cm)						
PREMENARCHEAL ^a								
Control	12	140.5	(1.8)	141.1	(1.9)	142.0	(1.9) ^b	
Train	12	142.2	(2.4)	143.7	(2.4)	144.2	(2.4) ^b	
POSTMENARCHEAL								
Control	20	163.2	(1.1)	163.4	(1.1)	163.5	(1.1)	
Train	21	162.7	(1.2)	162.9	(1.3)	162.9	(1.3)	

		SUM OF SKINFOLDS (mm)*						
PREMENARCHEAL								
Control	12	60.6	(7.1)	58.8	(16.9)	59.6	(7.6)	
Train	12	58.2	(9.3)	56.8	(3.4)	57.7	(8.9)	
POSTMENARCHEAL								
Control	20	67.4	(3.8)	67.5	(4.2)	65.1	(4.2)	
Train	21	76.6	(7.0)	76.6	(6.7)	78.3	(7.8)	

		PERCENT BODY FAT						
PREMENARCHEAL ^a								
Control	12	24.2	(6.1)	23.9	(5.7)	23.3	(6.2)	
Train	12	23.4	(6.0)	23.1	(5.8)	23.6	(5.6)	
POSTMENARCHEAL								
Control	20	26.6	(3.8)	26.5	(3.9)	26.0	(3.8)	
Train	21	27.6	(6.0)	27.7	(5.9)	26.2	(8.8)	

		LEAN BODY MASS (kg)						
PREMENARCHEAL ^a								
Control	12	27.1	(2.7)	27.7	(2.8)	27.8	(3.1) ^b	
Train	12	27.7	(5.3)	28.5	(5.3)	28.5	(5.3) ^b	
POSTMENARCHEAL								
Control	20	40.6	(3.8)	40.7	(3.7)	40.8	(3.8)	
Train	21	41.0	(5.3)	40.6	(5.7)	41.7	(5.0)	

* Note. Sum of triceps, subscapularis, biceps, iliac crest, medial calf.

^aPremenarcheal values significantly different from POSTM at all test periods.

^bSignificantly greater than PRETRAIN value.

4.1.5 Lean Body Mass

The PREM subjects had a lower mean lean body mass (LBM) than the POSTM subjects ($F(1,61)= 132.1, p <.001$). A main effect for Test was also found ($F(2,122)= 3.27, p <.05$). A secondary ANOVA, performed on the two Maturity groups separately, demonstrated that both PREM groups increased their LBM ($F(2,44)= 10.11, p <.001$) while LBM remained consistent over time for the POSTM subjects. As no Training level main effect was found, this increase in LBM cannot be attributed to the training protocol, but may reflect a growth effect.

4.2 Maximal Aerobic Power

Maximal aerobic power ($\dot{V}O_2 \text{ max}$) was described in four different ways: 1) in absolute terms ($\dot{V}O_2 \text{ max(abs), l} \cdot \text{ min}^{-1}$); 2) corrected for total body mass ($\dot{V}O_2 \text{ max(rel), ml} \cdot \text{ kg}^{-1} \cdot \text{ min}^{-1}$); 3) corrected for lean body mass ($\dot{V}O_2 \text{ max(lbm), ml} \cdot \text{ kg(lbm)}^{-1} \cdot \text{ min}^{-1}$); and 4) using height^{2.25} as a correction factor for differences in body dimensions ($\dot{V}O_2(\text{Ht(m)}^{2.25}, \text{ ml} \cdot \text{ m}^{-2.25} \cdot \text{ min}^{-1}$). All subjects attained a minimum of 2 criterion for $\dot{V}O_2 \text{ max}$ for each test period with the exception of one PREM-T subject (SS). The $\dot{V}O_2 \text{ max}$ and related data for this subject was not included in the following analyses. Table 4.2 provides the means and SE for the maximal aerobic power results of each group for the three testing periods.

When expressed in absolute terms, the PREM groups had significantly lower $\dot{V}O_2 \text{ max(abs)}$ scores at all test periods than the POSTM groups ($F(1,60)= 49.19, p <.001$). However, $\dot{V}O_2 \text{ max(rel)}$ was significantly higher in the PREM than the POSTM groups at each testing session ($F(1,60)= 9.73, p <.001$). Similar observations were made for the $\dot{V}O_2 \text{ max(lbm)}$ scores ($F(1,60)= 4.94, p <.05$).

Table 4.2:
Means and SE for Maximal Aerobic Power at Each Test for Premenarcheal and Postmenarcheal Subjects.

		$\dot{V}O_2 \text{ max } (l \cdot \text{min}^{-1})$					
		PRETRAIN	6wk		12wk		
	n						
PREMENARCHEAL^a							
Control	12	1.39 (.04)	1.50 (.06)	1.50 (.05) ^b			
Train	11	1.40 (.09)	1.48 (.09)	1.66 (.39) ^{bc}			
POSTMENARCHEAL							
Control	20	2.01 (.08)	1.97 (.10)	2.02 (.08)			
Train	21	2.06 (.08)	2.11 (.09)	2.23 (.07) ^{bc}			

		$\dot{V}O_2 \text{ max } (ml \cdot kg(\text{mass})^{-1} \cdot \text{min}^{-1})$					
		PRETRAIN	6wk		12wk		
	n						
PREMENARCHEAL^a							
Control	12	39.2 (1.6)	41.4 (1.9)	41.6 (1.7) ^b			
Train	11	38.7 (1.0)	40.3 (1.8)	44.3 (1.5) ^{bc}			
POSTMENARCHEAL							
Control	20	36.4 (1.3)	35.6 (1.7)	36.7 (1.5)			
Train	21	36.3 (1.0)	37.4 (1.1)	39.4 (1.1) ^{bc}			

		$\dot{V}O_2 \text{ max } (ml \cdot kg(\text{lbm})^{-1} \cdot \text{min}^{-1})$					
		PRETRAIN	6wk		12wk		
	n						
PREMENARCHEAL^a							
Control	12	51.7 (1.6)	54.5 (2.2)	54.4 (2.1) ^b			
Train	11	50.9 (1.2)	52.7 (1.9)	58.0 (1.3) ^{bc}			
POSTMENARCHEAL							
Control	20	49.6 (1.6)	48.4 (2.1)	49.5 (1.7)			
Train	21	50.1 (1.0)	51.9 (1.5)	53.7 (1.3) ^{bc}			

		$\dot{V}O_2 \text{ max } (l \cdot m^{-2.53} \cdot \text{min}^{-1})^*$					
		PRETRAIN	6wk		12wk		
	n						
PREMENARCHEAL							
Control	12	20.5 (0.6)	20.9 (0.9)	20.6 (1.0) ^b			
Train	11	20.1 (0.8)	20.8 (0.7)	23.0 (0.9) ^{bc}			
POSTMENARCHEAL							
Control	20	21.1 (0.6)	20.6 (0.8)	21.1 (0.7)			
Train	21	21.7 (0.6)	22.1 (0.8)	23.5 (0.6) ^{bc}			

*times 10⁴.

^aPremenarcheal values significantly different from POSTM.

^bSignificantly greater than PRETRAIN value.

^cSignificantly greater increase than in respective C group.

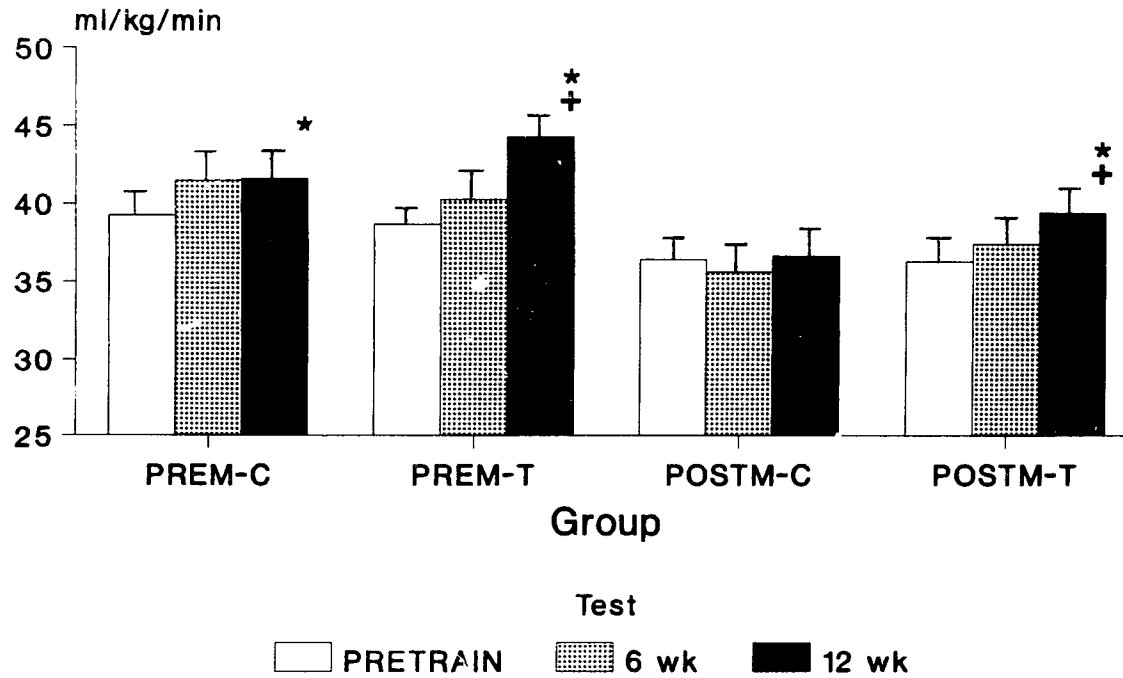
However, when $\dot{V}O_2$ max scores were expressed relative to Height^{2.25}, no significant difference was observed between the two maturity groups. Significant interactions between Maturity level and Test as well as between Training level and Test were also observed for each method of describing maximal aerobic power. Appendix I summarizes the ANOVA results of the preliminary analysis of maximal aerobic power.

The results of further analyses were similar for all methods of reporting maximal aerobic power. Therefore, the following description of results will refer to $\dot{V}O_2$ max(rel) (Figure 4.1).

Based on the Maturity by Test interaction for maximal aerobic power, the subjects were divided by Maturity level and differences in $\dot{V}O_2$ max between the T and C groups within each Maturity level were tested using a 2 x 3 ANOVA with repeated measures (Training level x Test). Both the PREM-T and PREM-C subjects significantly increased $\dot{V}O_2$ max ($F(1,21)= 16.72, p <.001$). However, a Training level by Test interaction showed that the 14.8% increase observed in $\dot{V}O_2$ max(rel) in the PREM-T exceeded the 6.2% rise in the PREM-C ($F(2,42)= 4.17, p <.01$). A similar interaction, found when differences in $\dot{V}O_2$ max were tested for within the POSTM subjects ($F(2,78)= 4.87, p <.01$), indicated that the POSTM-T subjects responded positively (8.7% increase) to the training program while the POSTM-C did not change (0.8% increase).

The training level by Test interaction found in the preliminary analysis supported further analysis of subjects separated by Training level. Thus, a second set of 2 x 3 ANOVA with repeated measures (Maturity x Test) was performed to test for differences between the two T groups and between the two C groups. The subtraction of the increase in relative $\dot{V}O_2$ max of the PREM-C (6.2%) from that

Maximal Aerobic Power (relative values)



- *Significantly different than PRETRAIN
- †Significantly greater than C group.

Figure 4.1: Means and SE for $\dot{V}O_2$ max(rel) at Each Test For Premenar-
cheal and Postmenarcheal Subjects.

of the PREM-T (14.8%) provided a means of reducing the influence that growth and development may have had on the $\dot{V}O_2$ max scores of the PREM-T group (8.6%). When the PREM-T and POSTM-T were compared, it was found that while the scores of the two groups were different from each other ($F(1,30)=21.56$, $p < .001$), both groups significantly improved their $\dot{V}O_2$ max over the duration of the study ($F(1,30)=21.13$, $p < .001$). The magnitude of this increase in $\dot{V}O_2$ max due to the training in the PREM-T group (8.6%) was similar to that in the POSTM-T group (8.7%). The lack of a Maturity by Test interaction supports the contention the both T groups responded to the training program in a similar way. This was a consistent finding regardless of the way $\dot{V}O_2$ max was reported.

Summary

To summarize, while both PREM groups significantly improved $\dot{V}O_2$ max, the increase observed in the PREM-T (14.8%) exceeded that seen in the PREM-C (6.2%). The actual training effect on the PREM-T was calculated as being the difference between the two PREM groups (8.6%). A training effect for $\dot{V}O_2$ max was observed in the POSTM-T subjects (8.7%), however the POSTM-C values remained constant (0.8%). The results demonstrate that the training effect for $\dot{V}O_2$ max was similar in the PREM-T and POSTM-T subjects.

4.3 Ventilatory Threshold

Ventilatory threshold (VT) was represented as the $\dot{V}O_2(\text{abs})$ at VT; the $\dot{V}O_2(\text{rel})$ at VT (VTrel); the $\dot{V}O_2(\text{lbm})$ at VT (VTlbm) and as a percentage of $\dot{V}O_2$ max(rel) (VT%) (Table 4.3). As with the $\dot{V}O_2$ max results, when VT was described in absolute terms (VTabs), the PREM scores were consistently lower than the POSTM

Table 4.3:
**Means and SE for Ventilatory Threshold (VT) at Each Test
 for Premenarcheal and Postmenarcheal Subjects.**

		VT(abs) ($l \cdot \text{min}^{-1}$)					
		PRETRAIN	6wk		12wk		
	n						
PREMENARCHEAL^a							
Control	12	1.05 (.06)	1.03 (.05)	1.01 (.08)			
Train	11	.91 (.08)	1.05 (.09)	1.12 (.07)			
PREMENARCHEAL							
Control	20	1.41 (.06)	1.35 (.07)	1.38 (.04)			
Train	21	1.56 (.09)	1.49 (.08)	1.47 (.06)			
		VT(rel) ($ml \cdot \text{kg}(\text{mass})^{-1} \cdot \text{min}^{-1}$)					
PREMENARCHEAL^a							
Control	12	29.2 (1.4)	28.8 (2.1)	28.2 (2.4)			
Train	11	25.6 (1.7)	28.2 (1.9)	30.2 (1.7)			
POSTMENARCHEAL							
Control	20	25.6 (1.0)	24.3 (1.4)	25.3 (1.0)			
Train	21	26.2 (1.1)	26.6 (1.1)	26.0 (1.1)			
		VT(lbm) ($ml \cdot \text{kg}(\text{lbm})^{-1} \cdot \text{min}^{-1}$)					
PREMENARCHEAL							
Control	12	38.8 (2.1)	37.8 (2.5)	36.8 (2.9)			
Train	11	33.3 (2.6)	37.0 (2.3)	39.6 (1.9)			
POSTMENARCHEAL							
Control	20	34.3 (1.3)	33.1 (1.7)	34.1 (1.2)			
Train	21	36.4 (1.7)	36.6 (1.7)	35.4 (1.3)			
		VT(% VO_2 max)					
PREMENARCHEAL							
Control	12	75.2 (4.0)	69.3 (3.4)	67.2 (4.1)			
Train	11	64.9 (3.9)	70.0 (3.5)	68.5 (3.6)			
POSTMENARCHEAL							
Control	20	70.5 (2.4)	68.2 (2.5)	68.9 (2.1)			
Train	21	72.1 (2.4)	70.9 (2.8)	66.5 (2.7)			

^aPremenarcheal values significantly different from postmenarcheal at all test periods.

scores ($F(1,60)=40.40, p < .001$). However, when expressed as $VT(\text{rel})$ the PREM scores were higher than the POSTM scores ($F(1,60)=5.44, p < .05$). No differences in either $VT(\text{lbm})$ or $VT\%$ between the two maturity groups existed. While the PREM-T subjects increased their VT by 24.37% ($SE=8.6$) the group means were not significantly different than the PREM-C scores. VT did not change for either of the POSTM groups over time. Figure 4.2 graphically displays the mean $VT(\text{rel})$ data for each of the groups at all test periods. No maturity related difference existed in the changes in VT by the T groups over the 12 wks. No differences between the maturity or training groups were observed when VT was expressed as $\% \dot{V}O_2$.

Summary

In summary, the PREM ventilatory thresholds were significantly different than the POSTM values except when expressed as a percent of $\dot{V}O_2 \text{ max}$ and as $VT(\text{lbm})$. No training effect was observed for either maturity group.

4.4 Maximal Heart Rates

Maximal heart rates (HR_{max}) were recorded to ensure that subjects exercised to exhaustion during each $VT/\dot{V}O_2 \text{ max}$ cycle test. HR_{max} did not change between tests confirming that subjects did exercise to exhaustion during each $VT/\dot{V}O_2 \text{ max}$ test. The only significant effect was the higher HR_{max} in the PREM-T group compared to the PREM-C ($F(1,21)=6.15, p < .05$) and both POSTM groups ($F(1,30)=19.42, p < .001$). This is primarily due to the max HR of subjects BL and SM (both in PREM-T) whose HR were consistent for all three tests at 217 and 219 $\text{b} \cdot \text{min}^{-1}$, respectively.

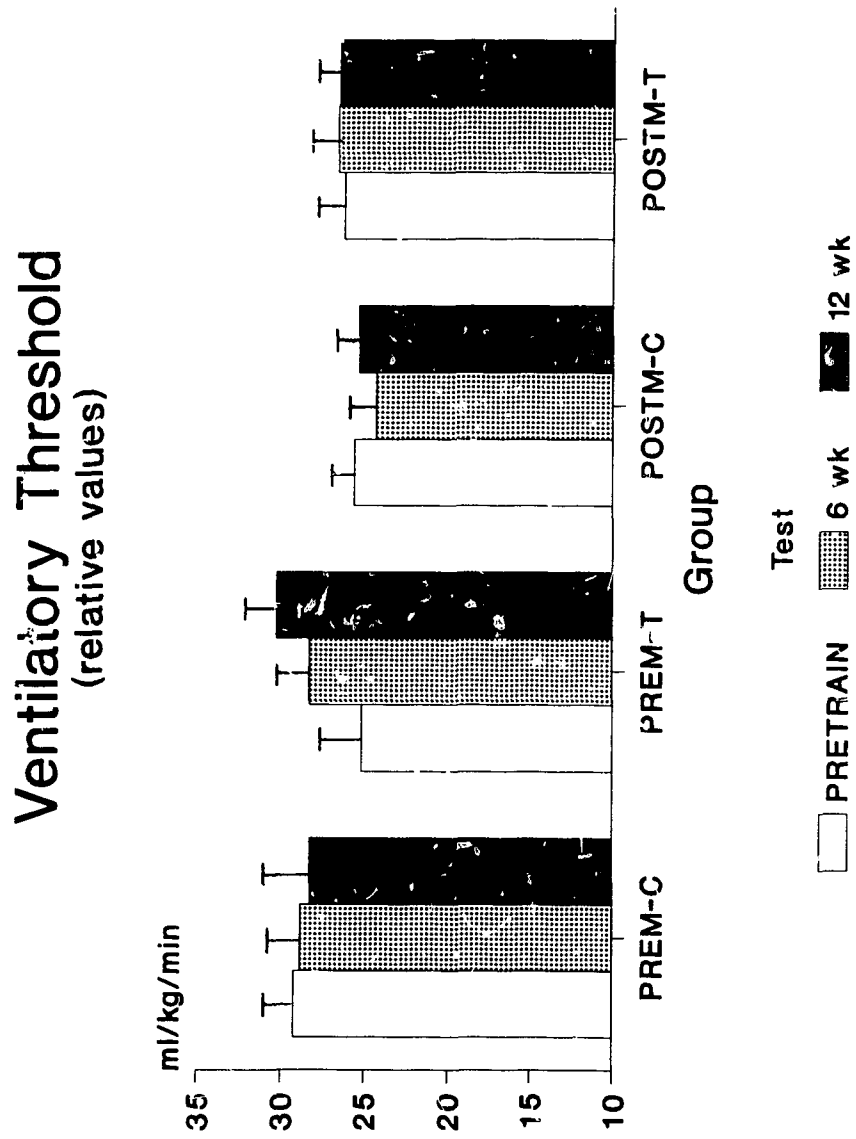


Figure 4.2: Means and SE for VT max(rel) at Each Test For Premenarcheal and Postmenarcheal Subjects.

4.5 Anaerobic Capacity

The total work performed during the 30s WAnt was used as an index of Anaerobic Capacity (AC). Similar to other performance variables, AC is a measure related to body mass. Means and SE for anaerobic capacity results for the three test periods are provided in Table 4.4. The results demonstrated that the PREM-T and PREM-C scores were significantly different throughout the study ($F(1,22)=5.02$, $p < .05$). Therefore, analysis of covariance with PRETRAIN scores as covariate was applied in order to correct for initial differences in AC. A 2 (Maturity) \times 2 (Training Level) \times 2 (Test) analysis of covariance (ANCOVA), with PRETRAIN score as the covariate, was used to test for differences in AC scores between the groups. All further description of AC refers to the ANCOVA results.

When expressed in absolute terms (ACabs; joules) the scores of PREM subjects were significantly lower than those of the POSTM groups ($F(1,60)=14.96$, $p < .001$). A similar finding was made when AC was expressed relative to lean body mass (ACl_{bm}; joules \cdot kg(lbm)⁻¹). However, when corrected for body mass (ACrel; joules \cdot kg(mass)⁻¹), no maturity difference was observed.

All groups demonstrated increases in AC(rel) overtime ($F(1,60)=4.54$, $p < .05$). Similar results were also obtained for AC(abs) and AC(lbm). The magnitude of change was the same for each group demonstrating that the training program did not exert an effect on the AC of either maturity group.

Summary

In summary, expressed relative to body mass, there were no differences in anaerobic capacity between the two maturity groups. While all groups demonstrated increases in AC, the rise observed in the training groups did not exceed

Table 4.4:
**Means and SE for Total Work Performed during the Wingate Test
 Reflecting Anaerobic Capacity (AC) for Premenarcheal and
 Postmenarcheal Subjects at Each Test.**

	n	AC(abs) (<i>Joules</i> · 10 ⁻²)		
		PRETRAIN	6wk	12wk
PREMENARCHEAL^a				
Control ^b	12	61.0 (3.2)	59.9 (2.7)	66.2 ^c (3.0)
Train	12	69.2 (4.8)	69.00 (5.8)	74.5 ^c (5.1)
POSTMENARCHEAL				
Control	20	102.3 (4.1)	102.2 (4.4)	114.3 ^c (3.6)
Train	21	100.9 (3.2)	102.1 (3.2)	112.3 ^c (2.7)
AC(rel) (<i>Joules</i> · kg(<i>mass</i>)⁻¹)				
PREMENARCHEAL^a				
Control ^b	12	170.2 (7.6)	163.8 (5.4)	182.5 ^c (6.6)
Train	12	189.7 (5.6)	183.7 (9.1)	197.5 ^c (4.8)
POSTMENARCHEAL				
Control	20	184.2 (5.0)	183.5 (6.0)	206.9 ^c (4.6)
Train	21	178.1 (4.4)	183.3 (6.1)	199.0 ^c (5.5)
AC(lbm) (<i>Joules</i> · kg(<i>lbm</i>)⁻¹)				
PREMENARCHEAL^a				
Control ^b	12	224.4 (7.7)	215.8 (7.1)	238.0 ^c (6.3)
Train	12	248.5 (8.2)	239.5 (11.5)	259.1 ^c (5.1)
POSTMENARCHEAL				
Control	20	251.3 (7.0)	250.1 (7.8)	279.8 ^c (5.7)
Train	21	253.3 (7.1)	253.3 (7.1)	271.0 ^c (6.3)

^aPremenarcheal values significantly different from postmenarcheal.

^bPremenarcheal C significantly different from premenarcheal T at all test periods.

^cSignificantly different from PRETRAIN.

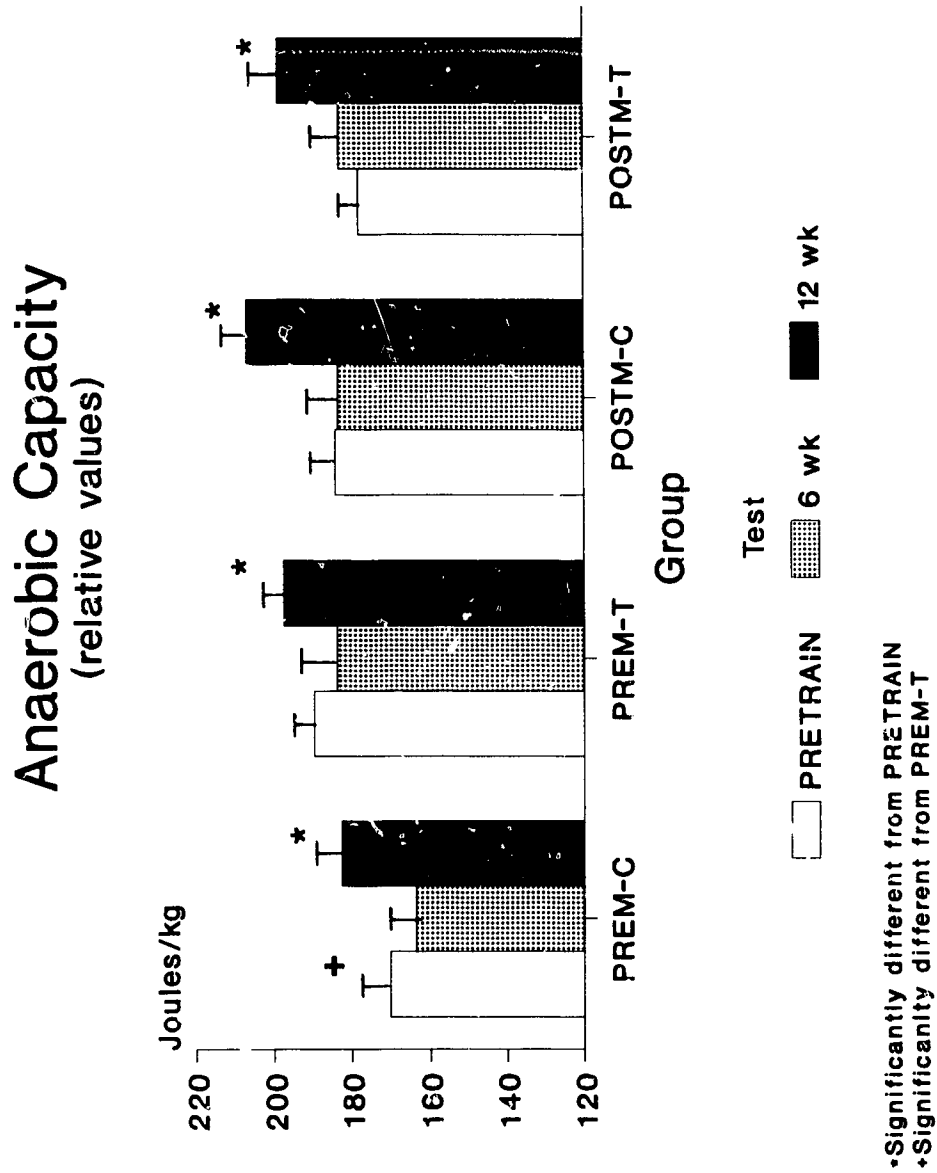


Figure 4.3: Means and SE for Total Work Performed During the Wingate Anaerobic Test Reflecting Anaerobic Capacity for Premenarcheal and Postmenarcheal Subjects at Each Test.

that of the control groups. Therefore, no training effect was achieved.

4.6 Blood Lipid Analysis

Blood samples from all subjects (N=65) were taken at both the PreTest and Post Test (at week 13) periods. However, due to sampling and analysis difficulties, some data are not available. For this reason, the subject numbers and the degrees of freedom (df) vary for each dependent variable as shown in Table 4.5 and Table 4.6. The greatest loss of data was in the analysis of the HDL-C sub-fractions (HDL₂ and HDL₃). This was the final serum analysis performed and in certain cases, not enough serum remained to be analysed appropriately. The large intersubject variability resulted in significant tests for homogeneity of variance between groups (BoxsM) ($p < .05$) for all lipid and lipoprotein variables with the exception of TC.

4.6.1 Total Triglycerides and Cholesterol

Total serum triglycerides (TG) and cholesterol (TC) levels remained consistent for each group. The means for TG and TC in PREM-T appeared to be consistently higher than any of the other groups, however these differences were not significant (Table 4.5).

Table 4.5:
Means and SE for Total Serum Triglyceride and Cholesterol
Concentrations for Premenarcheal and Postmenarcheal Subjects
Before and After the 12 week Training Program.

Group	n	Total Triglyceride ($mmol \cdot l^{-1}$)		Total Cholesterol ($mmol \cdot l^{-1}$)	
		PRETEST	POSTTEST	PRETEST	POSTTEST
PREMENARCHEAL					
Control	12	1.04 (0.10)	0.99 (0.09)	4.29 (0.25)	4.29 (0.16)
Train	12	1.33 (0.19)	1.23 (0.16)	4.40 (0.13)	4.29 (0.16)
POSTMENARCHEAL					
Control	20	0.85 (0.05)	0.91 (0.07)	4.08 (0.11)	4.11 (0.12)
Train	21	1.02 (0.10)	1.01 (0.10)	4.25 (0.15)	4.30 (0.17)

4.6.2 High Density Lipoprotein Cholesterol (HDL-C)

The HDL-C values, as measured by the PTA method (Chapter 3), did not differ between any of the groups at either test. No maturity or training effects were observed (Table 4.6).

4.6.3 HDL-C Subfractions

All HDL-C subfractions were determined from the HDL-C measured using the dextran sulfate method and therefore the sum of HDL₂-C and HDL₃-C varies slightly from the total HDL-C measured with the PTA method. Means and SE for the HDL₂-C and HDL₃-C subfraction concentrations are provided in Table 4.6.

HDL₂-C

While the serum concentrations of HDL₂-C decreased in all the groups from PRETEST to POSTTEST ($F(1,50) = 132.51, p < .001$), the drop in HDL₂ shown in Figure 4.4 was significantly greater in the PREM than in the POSTM groups ($F(1,50) = 26.73, p < .001$). Secondary analysis of the Training levels separately determined that this Maturity by Test interaction was due to the larger decrease in PREM-T than in POSTM-T subjects. ($F(1,26) = 37.82, p < .001$). However as no training effect existed in either of the Maturity groups this change in HDL₂ cannot be attributed to the training program.

HDL₃-C

All groups demonstrated significant increases in serum concentrations of HDL₃-C from initial levels ($F(1,51) = 12.22, p < .001$) (Figure 4.5). There was also evidence that this response was dependent on maturity status ($F(1,51) = 19.31$

Table 4.6:
Means and SE for High Density Lipoprotein Cholesterol (HDL-C)
Concentrations and the Subfractions HDL₂-C and HDL₃-C
for Premenarcheal and Postmenarcheal Subjects Before and
After the 12 week Training Program.

Group	n	Total HDL-C* (mmol · l ⁻¹)		n	HDL ₂ -C (mmol · l ⁻¹)		HDL ₃ -C (mmol · l ⁻¹)	
		PRE	POST		PRE	POST	PRE	POST
PREMENARCHEAL								
Control	12	1.41 (0.11)	1.31 (0.04)	9	0.66 ^a (0.13)	0.35 ^{abc} (0.08)	0.63 ^a (0.02)	0.90 ^{bc} (0.02)
Train	12	1.48 (0.10)	1.39 (0.09)	11	0.65 ^a (0.07)	0.26 ^{abd} (0.05)	0.65 ^a (0.03)	0.91 ^{bd} (0.03)
POSTMENARCHEAL								
Control	19	1.51 (0.05)	1.43 (0.06)	17	0.59 (0.05)	0.45 ^b (0.04)	0.80 (0.02)	0.90 ^b (0.02)
Train	21	1.49 (0.06)	1.49 (0.06)	18	0.53 (0.05)	0.41 ^b (0.06)	0.81 (0.02)	0.93 ^b (0.02)

*Phosphotungstic Acid method

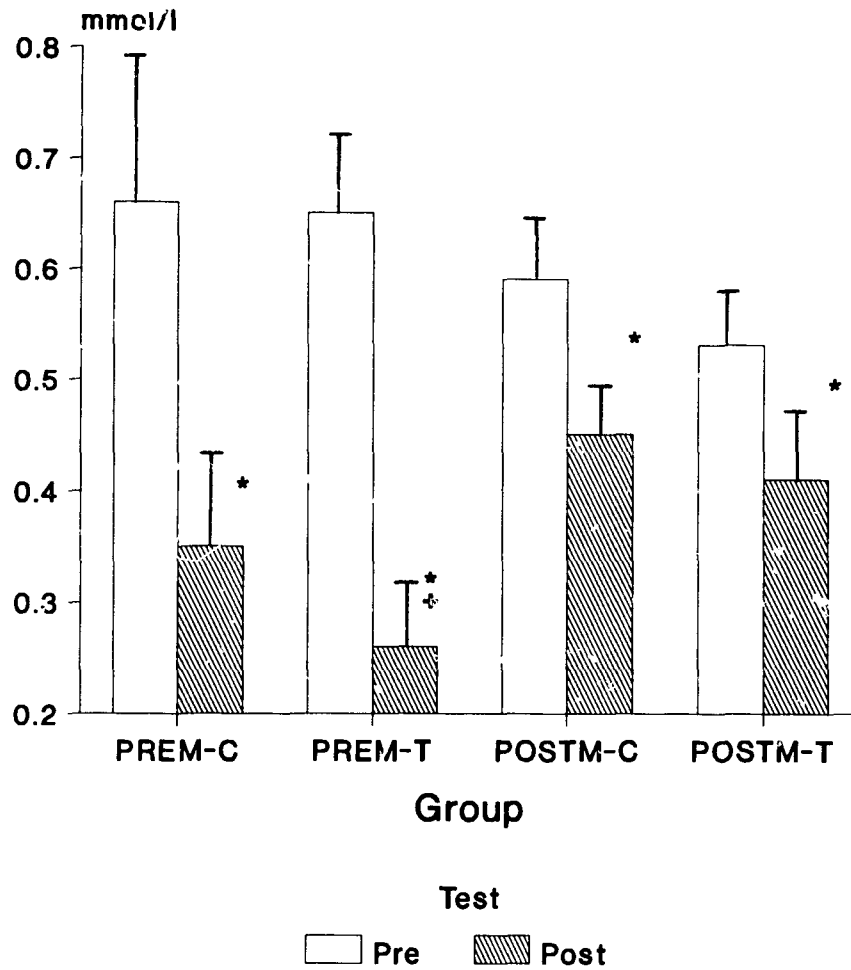
^aSignificantly different from postmenarcheal groups

^bSignificantly different from PRE score

^cSignificantly greater change than postmenarcheal-C

^dSignificantly greater change than postmenarcheal-T

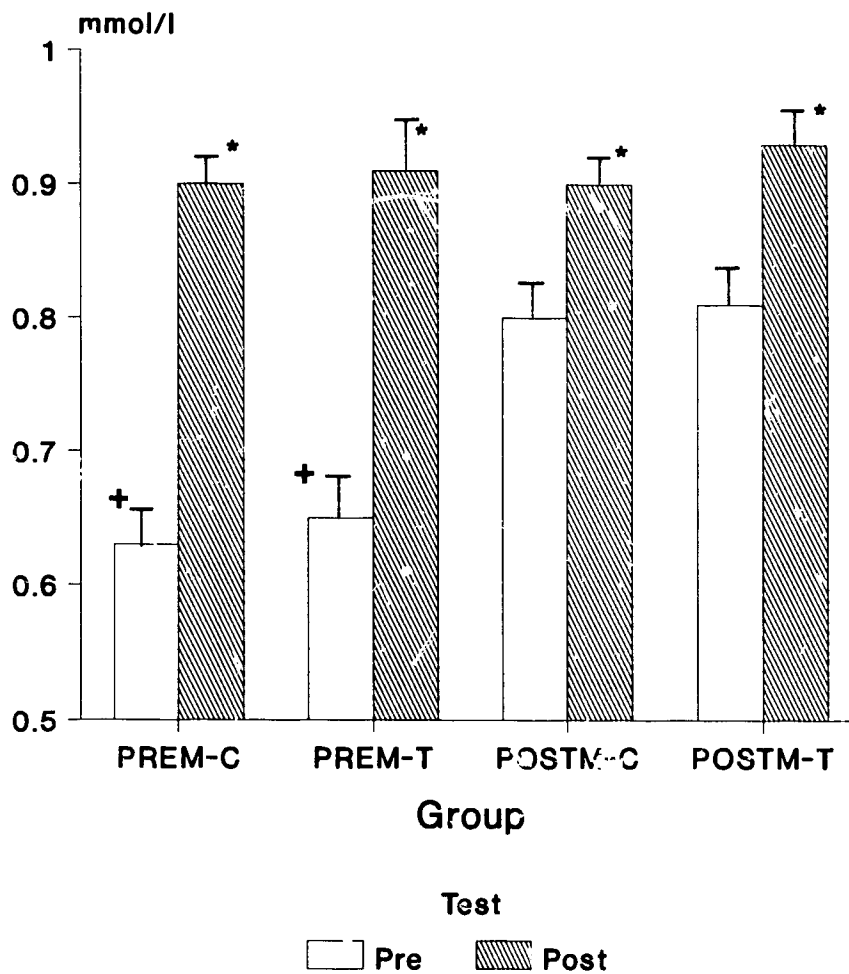
HDL₂-Cholesterol



*Significantly different from PRE
 **Significantly greater change than POSTM

Figure 4.4: Means and SE for PRE and POST HDL₂-C of Premenarcheal and Postmenarcheal Subjects at each test.

HDL 3-Cholesterol



+Significantly different from PRE score
 *Significantly lower than POSTM.

Figure 4.5: Means and SE for PRE and POST HDL₃-C of Premenarcheal and Postmenarcheal Subjects.

$p < .001$). By dividing the subjects according to Training level it was possible to locate and describe these differences. While both C groups increased their mean HDL₃-C concentrations ($F(2,48) = 49.01, p < .001$), a larger increase was observed in the PREM-C relative to the POSTM-C subjects ($F(2,48) = 10.14, p < .01$). When the two T groups were compared, results similar to that in the C groups were found. PREM-T and POSTM-T subjects each increased HDL₃ ($F(2,54) = 74.23, p < .001$), however similar to the PREM-C subjects, the mean increase in the PREM-T group was greater than that found in the POSTM-T ($F(2,54) = 8.8, p < .01$). No training effect existed for either of the Maturity levels. Therefore, these differences cannot be attributed to the training program.

4.6.4 HDL-C to Total Cholesterol Ratio

The lack of any alterations in TC or HDL-C resulted in no significant alterations to the HDL-C/TC ratio for any of the groups. In addition, no differences between any of the groups were observed.

4.6.5 Low Density and Very Low Density Lipoprotein Cholesterol

No significant differences between groups were observed in low density lipoprotein cholesterols (LDL-C) or very low density lipoprotein (VLDL) concentrations at either serum testing period. While the concentrations of LDL-C were slightly higher in the PREM subjects than the POSTM means, they were not statistically different (Table 4.7). There were no significant changes over time demonstrated by any group for either VLDL or LDL-C levels.

Table 4.7:
Means and SE for Low Density Lipoprotein Cholesterol (LDL-C)
and Very Low Density Lipoproteins (VLDL) Concentrations for
Premenarcheal and Postmenarcheal Subjects Before and After
the 12 week Training Program.

Group	n	LDL-C (<i>mmol · l⁻¹</i>)		VLDL (<i>mmol · l⁻¹</i>)	
		PRETEST	POSTTEST	PRETEST	POSTTEST
PREMENARCHEAL					
Control	12	2.66 (0.28)	2.78 (0.20)	0.47 (0.04)	0.45 (0.04)
Train	12	2.65 (0.14)	2.65 (0.17)	0.60 (0.08)	0.56 (0.07)
POSTMENARCHEAL					
Control	19	2.39 (0.11)	2.49 (0.12)	0.38 (0.02)	0.41 (0.03)
Train	21	2.56 (0.13)	2.60 (0.15)	0.46 (0.04)	0.45 (0.04)

Summary

To summarize, the training program did not elicit any changes in serum total triglycerides, total cholesterol, HDL-C, LDL-C, or VLDL. Since there were no changes in any of the lipids or lipoprotein cholesterols, there was also no alteration in the HDL-C to Total Cholesterol ratio. HDL₂-C were higher and HDL₃-C lower in the premenarcheal subjects than the postmenarcheal subjects. All groups demonstrated significant decreases in HDL₂-C and significant increases in HDL₃-C, however these changes were of greater magnitude in the premenarcheal subjects. No training effect was observed for the HDL subfractions in either maturity group.

4.7 Serum 17 β -Estradiol

Serum 17 β -Estradiol analysis revealed that all the PREM subjects were prepubertal. All subjects in this group had 17 β -Estradiol scores of less than 50 pmol·l⁻¹ at both serum test periods. The one exception to this (LM, PREM-C) had a PRETEST 17 β -Estradiol score of 100 mmol·l⁻¹ and POSTTEST value of less than 50 mmol·l⁻¹. These values fall below the rating of 'pubertal' according to published normal range values (B.C. Children's Hospital, 1987). All POSTM subjects had PRETEST and POSTTEST values above 120 pmol·l⁻¹ indicating that they were all within normal adult female 17 β -Estradiol range. PRETEST and POSTTEST values in the POSTM subjects varied according to the menstrual phase each subject was experiencing at the time of the blood collection. Individual blood 17 β -Estradiol results are provided in Appendix J.

4.8 Diet and Physical Activity Questionnaire

Analysis of the Diet and Physical Activity questionnaires, completed by all subjects at each test period, demonstrated that there were no obvious changes to regular dietary intake by any of the participants. Only one subject (GH; POSTM-T) reported being vegetarian and she remained so throughout the study. No subject was taking oral contraceptives at the time of the study and no one reported a smoking habit. Responses to the activity portion of the questionnaires indicated that habitual physical activity remained stable in the control group subjects throughout the study. The T group subjects reported no changes in their physical activity patterns with the exception of their participation in the training program three times a week for the 12 weeks of the study.

Chapter 5

DISCUSSION

5.1 Physical Characteristics

The 17β -estradiol levels of each premenarcheal subject fell into the category of pre-pubertal according to published standards (B.C. Children's Hospital) and are close to those reported for premenarcheal groups (Zonderland et al., 1984). The physical characteristics of both the premenarcheal and postmenarcheal subjects were similar to those reported for females of related ages and maturational level (Gilliam and Burke, 1978; Girondola et al., 1981; Peltenburg et al., 1984; Zonderland et al., 1984). Thus, the sample population was considered normal for young females of both maturity phases.

5.2 Training Effects on Anthropometric Variables

The training program did not elicit a significant effect on any of the anthropometric variables in either the premenarcheal or postmenarcheal groups (Table 4.1). The significant increases in mass and height reported in the premenarcheal subjects demonstrated the effects of normal growth during the period of the study. The increase in mass may be due to an increase in LBM rather than a change in %BF since only the fat free measure of LBM demonstrated a significant increase during the course of the study. Gains in height typically begin to decrease at approximately 12 years of age in females (Kemper and Verschuur, 1985; Rutenfranz

et al., 1984). The greater mean increase in height of the premenarcheal subjects as compared to the postmenarcheal subjects suggests that the latter group were probably closer to adult height than the less mature PREM subjects.

The large intersubject variation in sum of skinfolds was probably the reason for the lack of a maturity level main effect. The variability between subjects of similar developmental phases seen in the present study is similar to that reported in non-training control schoolgirls (Peltenburg et al., 1984).

The accuracy of estimating percent body fat (%BF) from anthropometric methods in pediatric populations is limited by problems related to the assessment of their validity. The results obtained by equations such as the one employed in this study (Durnin and Womersely, 1974) are at risk of yielding an overestimation of fatness (Boileau et al., 1985), especially for prepubescent subjects. Therefore, the comparison of %BF between the PREM and POSTM subjects is limited by the possibility of a greater overestimation occurring in the PREM than in the more mature POSTM subjects. The intent of this measurement was as a means of monitoring changes in body composition during the study. As the method remained constant throughout the study, comparisons of results from one test period to another for individual subjects, as well as subjects within a maturity group were considered appropriate. The significantly lower %BF observed in the PREM subjects in comparison to the POSTM subjects is similar to that reported by others (Kemper and Verschuur, 1985). If an overestimation in %BF did occur in the PREM subjects, a correction for this error would result in even greater differences than reported.

5.3 Maximal Aerobic Power

5.3.1 Maturation Differences in Maximal Aerobic Power.

The initial mean $\dot{V}O_2$ max of the premenarcheal and postmenarcheal subjects compare closely with those reported in the literature for young females of similar ages and maturity status (Bar-Or, 1983; Girondola et al., 1981; Mirwald and Bailey, 1986; Reybrouck et al., 1985; Rowland and Green, 1988; Rutenfranz et al., 1984; Weymans et al., 1985). The finding of a higher mean $\dot{V}O_2$ max (rel) in the premenarcheal subjects as compared to the postmenarcheal subjects (Table 4.2) support the results of others who have shown that relative maximal aerobic power declines in young females after approximately 13 years of age (Krahenbuhl et al., 1985; Rutenfranz et al., 1984). The most common explanation for this reported decrease in maximal aerobic power as females reach adolescence is the accumulation of subcutaneous fat during puberty (Bar-Or, 1983, p.4). This tissue type is metabolically inactive relative to muscle tissue and its accumulation results in an increase in total body mass with very little increase in metabolic activity.

Biologists have recognized that small animals exhibit higher oxygen uptake and greater heat production per unit body mass than larger animals (Taylor et al., 1970). As the surface area:mass ratio is greater in small animals compared to large animals, and heat is lost through surface area, the greater oxygen uptake per body mass reflects the metabolic survival adaptation used by warm-blooded animals to maintain internal body temperature. This Surface Law has been extended as a possible explanation for the differences in exercise oxygen consumption between females of different maturity levels (Rowland and Green, 1988). However, there is no empirical evidence to support the use of this theory to describe differences between subjects under the condition of exercise thermogenesis.

The standardization of maximal aerobic power based on growth-related differences in body dimensions and function has also been suggested (Bailey et al., 1978). The expression of $\dot{V}O_2$ max relative to height^{2.25} has been suggested as an alternative technique for correcting maximal aerobic power in order to compare the scores of individuals of different body size (Bailey et al., 1978; Mirwald and Bailey, 1986). In the present study, when height^{2.25} was applied to $\dot{V}O_2$ max in order to dissociate it from size, no differences between the PREM and POSTM subjects were found. These results support the findings of others who have suggested that height has an advantage over weight when used as the dimension by which maximal aerobic power is dissociated from size (Bailey et al., 1978; Mirwald and Bailey, 1986; Shephard et al., 1980).

There is some question regarding the most appropriate means by which $\dot{V}O_2$ max should be expressed when related to body dimensions. Height raised to the power of 2, 2.25, 2.46, and 3 have all been suggested (Bailey et al., 1978; Bar-Or, 1984; Krahenbuhl et al., 1985; Mirwald and Bailey, 1986; Shephard et al., 1980). In fact, Shephard et al. (1980) concluded that precise exponents may not be essential since, in their longitudinal study of children, $\dot{V}O_2$ max correlated equally well with height raised to a number of different values. The discrepancy regarding which exponent should be used in order to most accurately correct for differences between individuals has been attributed to quantitative, as well as qualitative, differences in the various physiological systems (Blimkie, 1989).

While the dimensional analysis method did eliminate any differences in $\dot{V}O_2$ max between the two maturity groups, there are a number of assumptions associated with it. The use of body dimensions to standardize physiological variables is based on the geometric similarity between individuals of varying size, and the stability of body tissue composition. The dimensional analysis theory assumes

that the relationships between size, proportion, composition and function remain constant across ages and gender (Bailey et al., 1978; Blimkie, 1989). In children, the composition of tissues has been reported to vary with age and sex (Lohman et al., 1984). Consequently, Blimkie (1989) has suggested that the application of these geometric principles to pediatric populations must be made with caution.

The oxygen uptake differences between children and adults can only partially be attributed to differences in body size and dimensions (Bar-Or, 1983, p.7). The higher $\dot{V}O_2$ max(rel) reported in prepubescent subjects compared to adults has been attributed to mechanical inefficiency rather than greater maximal aerobic power (Bar-Or, 1983:p8). However, as the mechanical efficiency of cycling is similar in children, adolescents, and adults (Bal, 1953; Bar-Or, 1983:p7), it is unlikely that this argument could be used to explain the higher relative $\dot{V}O_2$ max values in the PREM group. An alternative explanation could be that habitual activity patterns of the two maturity groups were different. However, precise quantification of habitual activities was not possible in the present study. Activity levels have been reported to decrease as females reach adolescence (Lenskyj, 1988). Reduced participation in aerobic activities could lead to an increase in adiposity and, consequently, a decrease in maximal aerobic power when expressed relative to body mass.

5.3.2 Effects of Training on Maximal Aerobic Power

Premenarcheal Subjects

The significant improvement in $\dot{V}O_2$ max in the trained premenarcheal subjects (PREM-T) exceeded that of the control group, demonstrating that $\dot{V}O_2$ max in the premenarcheal subjects was responsive to the aerobic training. The fact that

this finding was also observed when $\dot{V}O_2$ max was expressed relative to the size-dissociating factor of height^{2,25} provides additional evidence of a training effect in the PREM-T subjects. The increase in $\dot{V}O_2$ max found in the control premenarcheal (PREM-C) group, even though they did not participate in the training program, could be due to the effects of normal growth and maturation. Although it is possible that the habitual activity of this control group influenced their $\dot{V}O_2$ max, responses to the Physical Activity questionnaires indicated that there was no change in activity patterns of these subjects during study. There was an improvement in $\dot{V}O_2$ max in the PREM-C subjects however, the increase was not as great as that observed in the training group (6.2% versus 14.1%, respectively). As these subjects successfully attained $\dot{V}O_2$ max criteria at each test, the improvement was not due to initial scores being less than maximal. The reported decreases in maximal aerobic power in females with age have typically involved cross-sectional data for females over the age of 10 years. Morphological and physiological changes due to growth prior to puberty, such as myocardial size and function, may result in increases in $\dot{V}O_2$ max in children up until approximately 10–12 years of age. It is possible that the increased $\dot{V}O_2$ max values in the PREM-C subjects were the result of growth-related changes in cardiorespiratory function. In addition, since these subjects had not reached puberty, they had not yet experienced the typical increase in adiposity that often results in a decrease in relative maximal aerobic power in adolescent females.

The significant increase in maximal aerobic power of the PREM-T subjects during the 12 week training program is in contrast to the results of a number of studies. There has been considerable controversy regarding the trainability of pre-pubescent children, particularly those 10 years of age and under. Many

investigators have reported little or no change in $\dot{V}O_2$ max of prepubescents (Bar-Or, 1984; Bar-Or and Zwiren, 1973; Daniels and Oldridge, 1971; Daniels et al., 1978; Gatch and Byrd, 1979; Gilliam and Freedson, 1980; Kobayashi et al., 1978; Schmucker and Hollman, 1974; Stewart and Gutin, 1976; Yoshida et al., 1980). In many of these studies, improved running performances have been reported following training without any significant improvement of $\dot{V}O_2$ max. (Bar-Or and Zwiren, 1973; Daniels and Oldridge, 1971; Daniels et al., 1978; Kobayashi et al., 1978; Stewart and Gutin, 1976; Yoshida et al., 1980). The high relative oxygen cost of walking and running in prepubescent subjects compared to adults (Krahenbuhl et al., 1979; MacDougall et al., 1983) has been attributed to a metabolically expensive gait. The reported improvements in performance may simply reflect an increase in running efficiency due to the training program. However, as discussed earlier, this would not apply to cycling activities as the metabolic efficiency of cycling is similar for most individuals regardless of maturity status.

Reference to training program designs employed in such studies must be made before their results can be interpreted. Many of the above studies did not include adequate levels of exercise intensity, duration or frequency necessary to produce optimal training effects (Bar-Or and Zwiren, 1973; Gatch and Byrd, 1979; Stewart and Gutin, 1976). As reported in adults, in order for a training program to be effective the intensity and duration of the activity must exceed the levels that are normally experienced by the individual (ACSM, 1978). Several investigators have successfully increased $\dot{V}O_2$ max in prepubescent children provided that a training stimulus of greater than 60% $\dot{V}O_2$ max was incorporated in a program. (Brown et al., 1972; Docherty et al., 1987; Ekblom, 1969; Lussier and Buskirk, 1977; Mahon

and Vaccaro, 1989; Massicotte and MacNab, 1974; Rotstein et al., 1986; Vaccaro and Clarke, 1978). The incorporation of adult exercise criteria for intensity and duration has been proposed as the optimal method of successfully increasing maximal aerobic power in children (ACSM, 1988; Rowland, 1985; Vaccaro and Mahon, 1987). Pate and Blair (1978) concluded that improvements in $\dot{V}O_2$ max of prepubescent children were possible provided the program was a minimum of 12 weeks and included vigorous training intensity involving at least three sessions per week. The present study demonstrated that 12 weeks of continuous cycling three times a week at an intensity equivalent to approximately 65–70% $\dot{V}O_2$ max was sufficient to elicit improvements in maximal aerobic power in premenarcheal and postmenarcheal subjects.

The magnitude by which the trained premenarcheal subjects improved their $\dot{V}O_2$ max (8.6%) is consistent with the expectations of effective adult programs (ACSM, 1978) and is similar to the 7–15% rise reported in the pediatric exercise literature. While endurance exercise has been regarded as the optimal means of improving aerobic power (Rowland, 1985), Docherty et al. (1987) produced a significant 18% increase in $\dot{V}O_2$ max of young boys following 4 weeks of interval type training involving high intensity, short duration work. Similar findings have recently been reported by Mahon and Vaccaro (1989) who used a combination of steady state running and interval training with young 10–14 year old males.

Of the studies which have successfully produced increases in maximal aerobic power in children, very few have involved prepubescent females (Brown et al., 1972; Gilliam and Freedson, 1980; Lussier and Buskirk, 1977; Vaccaro and Clarke, 1978). The subjects involved in these studies often were a cross section of different maturity stages (Brown et al., 1972; Lussier and Buskirk, 1977). In

addition, two of these studies involved children who were participating in competitive athletic training at the time of the research and, thus, may not be considered representative of the general female prepubescent population (Brown et al., 1972; Vaccaro and Clarke, 1978). The subjects in the present study did not participate in any competitive athletic training prior to or during the present research and, as their initial aerobic characteristics demonstrated, were representative of the normal premenarcheal and postmenarcheal populations.

Postmenarcheal Subjects

Similar to that observed in the premenarcheal group, the trained postmenarcheal (POSTM-T) subjects exhibited a significant increase in $\dot{V}O_2$ max following the training program, while their maturity-matched controls (POSTM-C) demonstrated no change. No studies involving young females of similar age and maturity status are available with which to compare these results. As mentioned above, many studies have combined females of a wide range of ages and maturity status thereby making it impossible to separate the effects of training for children of different maturity levels.

The increase in $\dot{V}O_2$ max observed in the POSTM-T subjects is similar to that reported in pubescent and post-pubescent males (Eriksson and Koch, 1973; Mahon and Vaccaro, 1989; Weber et al., 1976). Cross sectional studies of boys of this age show that trained subjects exhibit higher $\dot{V}O_2$ max values compared to untrained peers (Daniels et al., 1978). As the influence of self-selection into these populations cannot be accounted for, interpretation of the results is difficult. When comparing the results of young males and females of similar age, maturity differences must be considered. Young females reach puberty up to two years

earlier than males (Marshall and Tanner, 1970) and therefore young males and females of similar chronological age may be at very different stages in maturity.

5.3.3 Maturation Effects on Training Response

In order to compare the effects of training on children of different levels of maturation, it is necessary to equate the conditioning dosage rather than apply similar absolute training loads to all subjects (Bar-Or, 1984). This was the case in the present study, as training intensity was determined as a percentage of the maximal heart rate for each individual subject. By equating the intensity of the training for all subjects based on a percentage of maximal heart rate, it was possible to compare the effects of training on the two maturity groups.

In summary, the results of this study demonstrated that both premenarcheal and postmenarcheal subjects successfully increased their maximal aerobic power with the endurance training and that the magnitude of this response was similar for each maturity group. These findings do not support the suggestion that up until 10–12 years of age training is not as effective as it is at older ages (Bar-Or, 1983; Ilmarinen and Rutenfranz, 1980; Yoshida et al., 1980). In addition, the similarity of training response observed in $\dot{V}O_2$ max for each maturity group suggests that maturity level has little influence on aerobic training effects on maximal aerobic power as suggested by others (Gilliam and Freedson, 1980).

5.4 Ventilatory Threshold

5.4.1 Maturation Differences in Ventilatory Threshold

The pretraining means for ventilatory threshold (VT) in the present study are similar to those reported by others for young females (Reybrouck et al., 1985;

Wolfe et al., 1986; Weymans et al., 1985). Anaerobic processes have been described as being lower in adolescents than adults (Bar-Or, 1984). Therefore, the higher VT(rel) in the premenarcheal compared to the postmenarcheal subjects may be due to a delay in the participation of anaerobic energy production (Table 4.3). An alternative explanation for the differences in VT between the maturity groups could be that the habitual activity patterns of the PREM subjects influenced aerobic capacity to a greater extent than the activity of the POSTM subjects. Similar findings were reported by Rowland and Green (1988), who reported greater mean VT(rel) values in premenarcheal subjects compared to adults females. In support of the findings by others (Mahon and Vaccaro, 1989; Weymans et al., 1985) all mean VT values, expressed as a percent of $\dot{V}O_2$ max, are higher than those reported in adults (Poole and Gaesser, 1986).

The mechanisms responsible for this maturity related difference have not been clearly defined. It has been observed that lower levels of the glycolytic rate-limiting enzyme, phosphofructokinase (PFK) and higher levels of the oxidative enzyme succinate dehydrogenase (SDH) exist in prepubertal and adolescent subjects (Eriksson, 1972; Haralambie, 1982). This metabolic enzyme profile has been implicated as a possible reason for lower anaerobic energy production in children and for their reliance on aerobic processes when performing physical work (Bar-Or, 1984).

The role of pubertal evolution for glycolytic enzymes in skeletal muscle of young males has been documented (Eriksson et al., 1971). However, more recent investigations have shown that glycolytic enzyme concentrations and activities in young males and females (Berg et al., 1986; Haralambie, 1982) may not be as low as originally reported (Eriksson et al., 1971; Eriksson, 1972). The inverse relationship between age and VT has been attributed to a rise in the capacity

for anaerobiosis as children reach puberty (Eriksson, 1980). Haralambie (1982) demonstrated that several enzymes involved in oxidative metabolism were higher in 13–15 year old males and females than in adults. The results were interpreted as evidence that the tricarboxylic acid (TCA) cycle functions at a higher rate in prepubertal and adolescent subjects compared to adults (Berg et al., 1986; Haralambie, 1982). It is possible that this is related to the low blood lactate levels reported in children (Eriksson, 1971) and the rapid onset of oxidative processes during exercise (Macek and Vavra, 1980). The higher concentration of oxidative enzymes relative to that in adults may explain the apparently greater reliance on aerobic processes by prepubescent and adolescent subjects. This would result in a delay in the requirement for anaerobic energy production during exercise of increasing intensity and therefore a higher VT.

5.4.2 Effects of Training on Ventilatory Threshold

The 24% increase in the VT of the PREM-T subjects was comparable to the improvement seen in 9–11 year old males following an eight week cycling program (Becker and Vaccaro, 1983). Similar to the present findings, the 25% increase in VT reported by Becker and Vaccaro (1983), was not significant when compared to untrained controls. In both studies, the large intersubject variation in VT may have been a contributing factor in the inability to demonstrate statistical changes. The lack of a training effect in VT for either maturity groups is in contrast with the findings of Mahon and Vaccaro (1989) who reported an increase in VT following an 8 week running program with young males. While these authors reported this increase in VT of their experimental group as being significant (19%), the rise observed in their control groups (5%) was not. These results were obtained

through individual comparisons of group means using multiple Student's t-test and at no time were the changes (measured as the difference between pre and posttests) in VT of the two groups compared.

While both the premenarcheal and postmenarcheal groups involved in the training program were able to increase their $\dot{V}O_2$ max, neither group demonstrated significant increases in VT. It is possible that the training intensity and protocol were not optimal for producing improvements in VT. However, VT has been reported to increase in adults following endurance training at an intensity equal to VT (MacDougall and Sale, 1981; Pool and Gaesser, 1985). While the intensity of the training used in this study was approximately equal to that of VT (65–70%), it did not appear to be appropriate to elicit any changes in VT. It may be that VT in young females is not as easily improved with training as it is in adults. Another possibility may be that young females require different training protocols than adults to elicit changes in VT. These results support the conclusions of Rotstein et al. (1986) who proposed that anaerobic threshold may not be as responsive as $\dot{V}O_2$ max following aerobic training in prepubescent and adolescent subjects. In addition, while Mahon and Vaccaro (1989) did report a significant increase in the VT of their 10–14 year old male subjects following an eight week training program, the increase was much smaller than changes observed in adults (Davis et al., 1979; Poole and Gaesser, 1985). An alternative explanation for the results of the present study may be that changes in VT would be difficult to obtain since the initial values were already high.

5.5 Anaerobic Capacity

5.5.1 Maturation Differences in Anaerobic Capacity

The mean anaerobic capacity (AC) scores, measured as the total work performed during the 30s Wingate Anaerobic test, are similar to values reported for prepubescent and adolescent females (Bar-Or, 1983; Grodjinovsky and Bar-Or, 1984; Tharp et al., 1984) and slightly lower than those for young males (Docherty et al., 1987; Grodjinovsky et al., 1980; Inbar and Bar-Or, 1986; Rotstein et al., 1986). The mean scores for the premenarcheal subjects were significantly lower than those of the postmenarcheal groups throughout the study (Table 4.4). An age-related difference in AC has been reported previously and may be attributed to qualitative changes to skeletal muscle rather than simply due to quantitative differences (Inbar and Bar-Or, 1986). The fact that correcting AC scores for differences in lean body mass did not diminish the difference between the two maturity groups support this contention. While ATP and CP concentrations are similar in young males and adults, the muscle content of glycogen is lower (Eriksson, 1972). Thus, anaerobic energy supply is restricted in children and may be a factor responsible for the low anaerobic capacity reported in pediatric populations (Bar-Or, 1984).

5.5.2 Effects of Training on Anaerobic Capacity

While all groups demonstrated significant increases in AC over the period of the study, the changes cannot be attributed to the 12 week endurance cycling program. In the only available study that considered the potential effects of exercise on AC in female children, Grodjinovsky and Bar-Or (1984) demonstrated higher AC scores in young females participating in a special sports class when

compared to controls who were involved in regular physical education classes. As their study did not involve a controlled training program, it is difficult to make comparisons with the results of the present research. Increases in AC following high intensity interval type training have been reported for young males (Rotstein et al., 1986; Grodjinovsky and Bar-Or, 1984; Mosher et al., 1985). However, a four week high intensity, short duration exercise program involving a combination of cycling and resistance training did not produce any improvements in AC of active young boys (Docherty et al., 1987). The inability to produce changes in AC in these subjects may have been due to their involvement in anaerobic type competitive sport just prior to the study which could have had the effect of reducing the responsiveness of the subjects to the training program.

Bar-Or (1984), suggested that an improvement of an initially low anaerobic capacity could account for the enhanced aerobic performances reported by others following training even though $\dot{V}O_2$ max did not improve (Bar-Or and Zwiren, 1973; Daniels and Oldridge, 1971; Daniels et al., 1978; Kobayashi et al., 1978; Stewart and Gutin, 1976; Yoshida et al., 1980). It was hypothesized that an improved anaerobic capacity with training could provide children with the ability to work closer to their $\dot{V}O_2$ max (Bar-Or, 1984). The H^+ formed with anaerobic metabolism would improve O_2 unloading from arterial blood and enhance O_2 uptake by the working muscles. The findings in the present study do not support this theory as the significant increases observed in $\dot{V}O_2$ max for both the premenarcheal and postmenarcheal subjects were attained with no concomitant training-induced improvements in anaerobic capacity.

5.6 Serum Total Triglycerides

5.6.1 Maturational Differences in Total Triglycerides

Mean serum total triglycerides (TG) values for all groups are similar to those reported in the Bogalusa Heart Study and are just above the published 50th percentile for females of similar age groups (Cresanta et al., 1984). In addition, the values agree with those reported by others for young females of similar maturity status (Nizankowska-Blaz and Abramowicz, 1978; Valimaki et al., 1980). Maturity status did not appear to have an effect on the serum levels of TG.

5.6.2 Effects of Exercise on Total Triglycerides

The training program implemented in this study did not elicit any significant changes in TG for either the premenarcheal or postmenarcheal subjects (Table 4.5). Lower serum TG concentrations have been reported in active pre-adolescent children as compared to inactive children (Bell et al., 1989; Thorland and Gilliam, 1981). However, no studies that have involved training with children have demonstrated an exercise-induced effect on TG. From longitudinal studies of adults, it is apparent that a significant decrease in TG occurs with training only if initial, pretraining levels are elevated (Haskell, 1984). This may explain the stable TG results as no subject demonstrated abnormally high TG levels at anytime during the study.

5.7 Serum Total Cholesterol

5.7.1 Maturational Differences in Total Cholesterol

Mean serum total cholesterol (TC) concentrations agree closely to values for young females of similar maturity status (Nizankowska-Blaz and Abramowicz, 1978; Valimaki et al., 1980; Zonderland et al., 1984) and are just above the 50th percentile reported by the longitudinal Bogalusa Heart Study for female children of similar ages (Cresanta et al., 1984). No differences in TC were exhibited between the two maturity groups.

5.7.2 Effects of Exercise on Total Cholesterol

The training program did not produce changes in TC for either the premenarcheal or postmenarcheal subjects (Table 4.5). This finding is similar to other pediatric exercise studies where TC remained consistent following endurance type training (Gilliam and Burke, 1978; Linder et al., 1983; Savage et al., 1986). In studies of adults, TC levels have been significantly reduced through the participation in exercise programs (Paffenbarger et al., 1978). However, initial levels have generally been high in those subjects who demonstrated significant decreases in TC. In addition, the duration of training programs which have effectively altered TC in adults have typically been greater than three months (Williams et al., 1982). In the present study, the highest TC values were observed in two subjects (JH, PREM-C; JG, POSTM-T) who demonstrated TC values just above the 75th percentile for females of similar ages (Cresanta et al., 1984). However, these levels remained stable for both subjects across tests.

Exercise has been shown to have a direct influence on the control of atherosclerosis by altering lipoprotein cholesterol profiles (Haskell et al., 1980; Wood and

Haskell, 1979). While cholesterol levels appear to be related to atherogenic development, it is the measurement of each of the lipoprotein bound cholesterols which gives a more precise description of the apparent relationship between exercise and coronary heart disease (CHD) risk than does the measurement of TC (Haskell, 1980).

5.8 Serum Lipoproteins

A consistent serum concentration of TC may not reflect stable proportions of cholesterol bound to the various lipoproteins. Therefore it is necessary to assess the serum content of each lipoprotein in order to evaluate the influence of intervention methods on the lipoprotein profiles. Of primary importance is the effect of exercise on HDL-C, as it has been shown to have strong anti-atherogenic properties (Wood et al., 1984). Although LDL-C concentrations have been related to increased coronary heart disease risk the effects of exercise on this lipoprotein do not seem to be as great as those on HDL-C.

5.8.1 Maturational Differences in Lipoprotein Cholesterols

The mean serum HDL-C and LDL-C concentrations observed in the premenarcheal subjects were similar to those of the postmenarcheal subjects (Table 4.6; Table 4.7). The values are in agreement with previously published values for young females (Gilliam and Burke, 1978; Valimaki et al., 1980; Zonderland et al., 1984) and are within the 50th percentile of the Bogalusa Heart Study for females of similar ages (Cresanta et al., 1984). The mean HDL-C values are close to (Savage et al., 1986) or slightly lower (Thorland and Gilliam, 1981; Valimaki et al., 1980) than those reported for young males.

5.8.2 Effects of Exercise on Lipoprotein Cholesterols

Results from studies of adults have demonstrated that endurance exercise can elevate HDL-C levels provided the training program was of a sufficient intensity and duration (Ballantyne et al., 1982; Peltonen et al., 1981; Wood et al., 1984). HDL-C levels have been reported to be higher in active, fit adults than in sedentary individuals (Dufaux et al., 1982; Durstine et al., 1987; Nikkila et al., 1978; Wood and Haskell, 1979). There is little empirical data available to indicate if exercise has a similar effect on HDL-C in children. As the process of atherosclerosis begins in childhood (Kannel and Dawber, 1978) it is necessary to identify factors that may influence this process.

No training effects were observed in HDL-C, LDL-C or VLDL in the premenarcheal or postmenarcheal subjects. There are few data concerning the effects of exercise training on the serum lipoproteins of children against which these findings can be compared. Most studies have not involved any systematic training program but rather have been limited to comparisons between highly active and inactive groups (Bell et al., 1989; DuRant et al., 1982; Nizankowska-Blaz and Abramowicz, 1978; Parizkova et al., 1986; Thorland and Gilliam, 1981; Valimaki et al., 1980; Zonderland et al., 1984). Results from some of these studies have demonstrated higher HDL-C and lower LDL-C levels in active subjects compared to children who are less active (Bell et al., 1989; DuRant et al., 1982; Nizankowska-Blaz and Abramowicz, 1978; Valimaki et al., 1980; Zonderland et al., 1984). However, others have failed to show any differences in HDL-C or in LDL-C between the two groups (Thorland and Gilliam, 1981). The findings of studies using exercise as a means of changing blood lipoprotein levels in children have been equivocal. Difficulties interpreting the results of these studies stem from research design

problems including lack of non-training controls and inadequate exercise design and description (Deveaux et al., 1986; Gilliam and Burke, 1978; Linder et al., 1983).

Several longitudinal studies unable to show significant increases in HDL-C in adult females, concluded that HDL-C in women may be less responsive to exercise training than in males (Allison et al., 1981; Ballantyne et al., 1992; Brownell et al., 1982; Frey et al., 1982; Lipson et al., 1980). A higher HDL-C concentration in adult females compared to males has been attributed to the overriding effects of endogenous estrogens (Beaglehole et al., 1980). However, this protective effect could only be applicable to the postmenarcheal subjects in the present study as the sex hormones play no determining role on serum lipids or lipoproteins prior to menarche (Zonderland et al., 1986). While the HDL-C values for the subjects in this study were not higher than those reported for young males of similar age (Thorland and Gilliam, 1981; Savage et al., 1986; Valimaki et al., 1980), gender-related comparisons are difficult to make as the maturation of the male subjects were not clearly defined.

Similar to the findings of the present study, a number of investigations of adults and children have reported little or no change in serum lipids and lipoproteins even though the exercise training was sufficient to improve the aerobic conditioning of the subjects (Frey et al., 1982; Linder et al., 1983; Ready and Quinney, 1982; Savage et al., 1986; Williams et al., 1982). A dose-response relationship between activity intensity and lipoproteins has been proposed (Powell et al., 1987). Based on a number of longitudinal studies of adults, it has been suggested that a minimum of three months, and perhaps as much as a full year, is necessary before changes will occur (Pate and Blair, 1978; Williams et al., 1982; Wood et al., 1984). To date, investigations of the possible effect of exercise intensity provide

no conclusive evidence of an intensity threshold existing for changes in blood lipid profiles (Gaesser and Rich, 1984; Savage et al., 1986).

No long term studies have been attempted with children. In fact, the 12 week duration of the present training study exceeds all other pediatric exercise studies designed to evaluate the effects on serum lipids and lipoproteins of healthy children. The results of this study demonstrate that 12 weeks of aerobic training did not produce an effect on blood lipid profiles in healthy children. Whether or not an increase in the duration of training will result in significant alterations to blood lipid profiles remains to be determined. Gilliam and Burke (1978), reported significant increases in HDL-C concentrations of prepubertal females after only 6 weeks of aerobic training. However, interpretation of their results is limited due to the lack of an untrained control group against which the values of the training groups could be compared.

Savage et al. (1986) speculated that HDL-C values in their young male subjects would have ultimately increased if the training had been extended for longer than the reported 10 week walk/jog program in their study. a longer duration, HDL-C ultimately would have increased (Savage et al., 1986). In consideration of the longitudinal findings from studies of adults, it is possible that the exercise training in the present study was not long enough to elicit such changes. It has been suggested that a critical level for the lipoproteins, especially HDL-C, may exist in children, above which the influence of exercise is limited (Bell et al., 1989; Linder et al., 1983). Therefore the stability of the blood lipids and lipoproteins following the exercise training may be explained by the normal values in the young females prior to the study. Perhaps blood lipid profiles in children are only responsive to exercise if they are initially abnormal and that little change should be expected if normal levels already exist.

Diet has been shown to affect HDL-C and TC levels (Heiss et al., 1980). Analysis of the Dietary questionnaire indicated intrasubject consistency for dietary intake throughout the study. However, these results must be interpreted in light of the limitations of this recall method. Smoking, alcohol consumption (Elkeles et al., 1983; Stubbe et al., 1982) and oral contraceptives (Havekes et al., 1981) have each been implicated as factors influencing serum lipid and lipoproteins, particularly HDL-C. However, based on information collected from the diet and physical activity questionnaire, it was determined that these factors had no influence on the premenarcheal and postmenarcheal subjects in this study.

5.9 HDL-C Subfractions

5.9.1 Maturational Differences in HDL₂-C and HDL₃-C

A unique aspect of this study was the inclusion of the HDL-C subfractions, HDL₂-C and HDL₃-C, in the assessment of the effects of exercise training on young females. Premenarcheal subjects had higher HDL₂ and lower HDL₃ concentrations than the postmenarcheal subjects (Table 4.6). In adult males, the concentration of HDL₂-C is normally one third that of HDL₃-C values (Gidez and Eder, 1984). However, Durstine et al. (1987) reported HDL₂-C values that were very close to HDL₃-C levels in elite women distance runners. The results of the present study demonstrated that the HDL₂-C concentration was initially equal to HDL₃-C in the premenarcheal subjects, but by the end of the study had dropped to approximately one third of the HDL₃-C level. HDL₂-C remained approximately one half that of HDL₃-C in the postmenarcheal subjects for each test.

5.9.2 Effects of Exercise on the HDL-C Subfractions

As each training and control group exhibited significant decreases in HDL₂-C and increases in HDL₃-C, it is not possible to attribute the changes to the training program. It is unlikely that the alterations in the HDL-C subfractions were due to dietary factors as the other lipid levels remained constant throughout the study. It would appear that the larger magnitude of change in both HDL₂-C and HDL₃-C in the premenarcheal compared to the postmenarcheal subjects may be a maturity-related phenomenon. A greater variability in these measures in less mature subjects may be a possible explanation for the differences. In adults, exercise has been shown to increase HDL₂-C while HDL₃-C remains constant (Krauss et al., 1979; Wood and Haskell, 1979; Durstine et al., 1987).

Durstine et al. (1987) showed higher HDL₂-C and lower HDL₃-C levels in the elite women runners in contrast with sedentary controls. However, as this was not a training study, interpretation of these differences in the concentration of HDL-C subfractions is limited. To date, no pediatric exercise research involving the study of HDL₂-C and HDL₃-C are available. Zonderland et al. (1984) reported higher apoprotein A-1 (Apo A-1) levels in endurance trained premenarcheal swimmers compared to that of untrained controls. Apo A-1, the major protein of HDL, acts as a cofactor for LCAT resulting in the esterification and transfer of cholesterol from peripheral tissues to the HDL particle (Dufaux et al., 1982). The proportion of Apo A-1 in HDL₂ is greater than in HDL₃ (Eisenberg, 1984). While increases in Apo A-1 have not been directly related to increases in HDL₂, such a relationship may eventually be demonstrated.

To summarize, the 12 week endurance training did not elicit any significant

changes in the blood lipids or lipoproteins in either the premenarcheal or postmenarcheal subjects. A number of possible explanations for this lack of change can be made. It is possible that the duration of the program was not sufficient to produce changes in the lipids and lipoproteins in the trained subjects even though improvements in maximal aerobic power occurred. However, as no other study has demonstrated significant changes in children compared to untrained controls, it is likely that the responsiveness of lipids and lipoproteins is dependent upon initial levels. It is possible that the blood lipid profiles in the subjects of this study were not responsive to the training since they were already at normal levels.

5.10 Conclusions

The specific findings of this study permit the following conclusions to be drawn:

1. The training program did not influence any measure of body composition in either the premenarcheal or postmenarcheal subjects. Since any change that did occur in the premenarcheal subjects can be attributed to normal growth and maturational effects H_0-1 cannot be rejected. As no training effects were observed for either maturity group, hypothesis H_0-5 also cannot be rejected.
2. $\dot{V}O_2$ max is sensitive to endurance exercise training in premenarcheal subjects as the increase in maximal aerobic power in the PREM-T subjects was significantly greater than that seen in the control group. This increase was of a similar magnitude as the increase reported for the postmenarcheal training group. It is suggested that the response of $\dot{V}O_2$ max in young females is not affected by maturity status. Thus, null hypothesis H_0-2a , stating that exercise training would not effect the cardiorespiratory variable of maximal

aerobic power, must be rejected. However, as no maturity difference existed regarding the responsiveness of $\dot{V}O_2$ max, H_o -6a is not rejected. It can therefore be concluded that young females should be encouraged to participate in aerobic endurance type activities in order to improve and maintain cardiorespiratory health and fitness.

3. With regards to aerobic capacity, the exercise program did not significantly change ventilatory threshold (VT) in either the premenarcheal or postmenarcheal groups. The results of the present study provide no evidence of a training effect existing for VT, therefore H_o -2b cannot be rejected. As neither maturity group demonstrated significant changes in VT hypothesis H_o -6b, which states that there would be no maturity difference in response of VT to the training, must also not be rejected.
4. The increases in maximal aerobic power reported in both the premenarcheal and postmenarcheal subjects following the training occurred without any concomitant improvement in anaerobic capacity (AC). Therefore, hypothesis H_o -3 is not rejected as none of the training groups exhibited increases in AC exceeding that observed in the control group following the exercise program. Hypothesis H_o -7 is also not rejected since the premenarcheal and postmenarcheal AC scores were both stable over time.
5. Serum total triglycerides, total cholesterol, HDL-C, LDL-C and VLDL concentrations were not affected by the 12 week training program in both the premenarcheal and postmenarcheal subjects. Therefore, hypothesis H_o -4a and H_o -4b, referring to the inability of the blood lipids and lipoproteins to respond to training, are not rejected. It is also concluded that while the HDL-C subfractions were not as stable in the premenarcheal subjects when

compared to postmenarcheal subjects, HDL₂-C and HDL₃-C concentrations in neither maturity group were affected by the training program. Therefore, hypotheses H_o-4c is not rejected. Since no training-induced alteration in the blood lipids, lipoproteins or HDL-C subfractions were elicited in either maturity group, hypotheses H_o-8a, H_o-8b, and H_o-8c must not be rejected.

5.11 Directions for Future Research

The findings of this pediatric exercise training study suggest the following directions for future research.

1. To more fully assess the influence maturation has on the responsiveness to exercise, future research should incorporate a longitudinal training program that would monitor the responsiveness of young females to exercise as they mature. Longitudinal studies of circumpubertal males have reported maturity related changes in aerobic parameters (Cunningham et al., 1984; Mirwald et al., 1981), but have not involved any controlled exercise training in their research designs. A longitudinal study of circumpubertal females has yet to be conducted. Such an approach could provide corroborative support to the findings of the present study.
2. The lack of change in ventilatory threshold following the 12 weeks of training suggests that the relationship between type of training and the various measurements used to reflect aerobic fitness be investigated. The incorporation of interval, as well as continuous training at different intensities, into a research design will provide a more clear understanding of aerobic exercise and changes in cardiorespiratory health and fitness.
3. It is recommended that research involving young males of varied maturity levels be carried out to examine whether or not differences in responsiveness to exercise exist for this gender. This approach might provide greater confidence in the present finding that maturity in young females does not influence the ability to respond to endurance exercise.

4. A limitation of this study was the reliance on personal recall of diet and activity patterns. Therefore, it is advised that further research in the area of exercise and children involve greater investigator control and daily monitoring of dietary intake and habitual physical activity of subjects.
5. Investigators in the field of exercise and pediatric blood lipid profiles should consider including the evaluation of the apolipoproteins. These protein structures of the lipoproteins have recently been suggested as being more sensitive than HDL-C or LDL-C to exercise (Zonderland, 1985). In addition, the concept of a dose-response relationship for the blood lipids and lipoproteins in young males and females should be incorporated into future pediatric research. Further investigation is required before any conclusive statement regarding the role of exercise on blood lipid profiles in young females can be made.
6. The often inconclusive and controversial results in pediatric exercise literature suggest that more research be conducted in the study of exercise as a means of reducing coronary heart disease (CHD) risk factors in children. A limitation of this study was the reliance on the blood lipid and lipoprotein cholesterol as indices of CHD risk. The incorporation of other risk factors such as obesity, blood pressure, and family history in future research is recommended. In addition, further investigations of larger groups of children, especially females and children with abnormal blood lipid profiles, are needed before any conclusive statement regarding the role of exercise on blood lipids and lipoproteins in pediatric populations, and its prophylactic influence on the development of atherosclerosis in later life, can be made.

7. Finally, the young female subjects of this study demonstrated that, when provided with the experience, they were very motivated to participate in regular aerobic training. The participants of both T and U groups have demonstrated increased awareness and participation in physical activities since the conclusion of this study. However, this evidence is strictly anecdotal. Future research in the field of pediatric exercise physiology should include some method of assessing changes in behaviors and attitudes towards exercise.

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Appendix A

American College of Sport Medicine Position Statement on Children and Exercise

“It is the opinion of the American College of Sport Medicine that physical fitness programs for children and youth be developed with the primary goal of encouraging the adoption of appropriate lifelong exercise behavior in order to develop and maintain sufficient physical fitness for adequate functional capacity and health enhancement.”

(ACSM, 1988)

Appendix B

Letter to Parents and Informed Consent

TO: Parents/Guardians of students interested in participating in the 'Exercise and The Young Female' research study.

FROM: Dr. David Docherty, Ph.D, Professor
Catherine A. Gaul, B.Ed, M.Sc.
School of Physical Education
University of Victoria

We are conducting a research project to determine how regular aerobic training effects the cardiovascular fitness of young females and how physical maturity influences a girl's responsiveness to exercise. We are also interested in testing how this type of exercise alters the blood cholesterol levels of young girls. Although there is considerable data from studies involving young boys, very little information regarding exercise and young females has been collected.

The girls participating in this study will be randomly assigned to one of two groups: 1) Training group or 2) Control group. The training group will be involved in a 12 week program consisting of 30 minutes of cycling on stationary bikes three times a week. The intensity of this exercise will be enough to raise the girls' heart rates to approximately 75% of maximum. This would be a heart rate of about 140-150 beats per minute which is no greater than what would be reached during normal child play. This exercise will take place in the school and will always be conducted under close supervision by the primary investigators (see above) and one other research assistant from the university. The training program will NOT interfere with the school day. Times for training will be organized in the morning before classes and in the afternoon after school.

The study will involve a number of exercise tests to be performed throughout the period of training. All girls will participate in these tests no matter which group they are in. There will be 4 tests in all: 2 pre-tests, a test at the end of week 6 of the training program and a post-test at the end of the study (week 12). There will be 4 sections to each test: 1) Body composition, 2) Aerobic fitness, 3)

Anaerobic capacity, 4) Blood analysis. All these tests will be conducted at the Sport and Fitness Center in Physical Education at the University of Victoria.

The tests to be performed include:

1. Body Composition

Selected body measurements will be taken to describe size and composition of each girl. These will include height, weight, 5 skinfold measures and some muscle and bone width measurements. The data collected from this section of tests will allow us to monitor any changes in the body during the period of the study.

2. Aerobic Fitness

Aerobic fitness will be determined through a progressive continuous test on the cycle ergometer. Expired air is collected through a mouthpiece and analyzed by a metabolic cart for oxygen content. This is the most precise method for measuring cardiovascular, or aerobic, fitness. Although it is a maximum test the intensity will not exceed the stress of vigorous play. Your daughter will be closely monitored throughout the test by trained personnel.

3. Anaerobic Capacity

The girls will be given a 2 minute warm up on the cycle ergometer before they begin this supramaximal test. The test involves a 30 second "all-out" performance on the cycle ergometer. The girls will be instructed to ride as fast as they can for 30 seconds. We are interested in finding out how much power they can produce and how well they can maintain this power during the 30 seconds of the test.

4. Blood Analysis

An important component of this study is the determination of the influence exercise has on blood lipid profiles. A 10ml blood sample will be taken from each girl on the test days by a laboratory technician with pediatric experience from Island Medical Laboratories. The procedure will be performed under strict sterile conditions. These blood samples will be analyzed for specific blood components: High density (HDL) and low density (LDL) lipid cholesterols, total cholesterol, and total triglycerides. These variables are useful in describing the risk of coronary heart disease. By monitoring them, we will be able to determine

the role exercise has in reducing this risk in young girls by altering their blood lipid profiles. In addition, the blood will be analyzed for certain hormones in order for sexual maturity to be assessed. As explained earlier, one of the objectives to the study is to determine if maturity level effects a girl's responsiveness to exercise. Therefore, it is necessary for us to assess maturity level. In total, there will be 3 blood samples taken over the course of the study: A pre-test sample, one at week 6, and one at the end of the study (week 12).

All the testers and trainers involved in this study are fully certified technicians from the Sport and Fitness Center at U.VIC. All data collected will be considered confidential in nature and will be used only for the present study. Upon completion of the project we will provide you with a confidential report of your daughters' results.

This study has been approved by the Committee on Research involving Human Subjects at the University of Victoria, the Superintendent of School District no. 61, and the principal of your school.

Each subject should understand that she is free to withdraw from the project at anytime. However, we would like to emphasize the importance of having as many subjects as possible complete the study.

We look forward to beginning this exciting project with the girls. Should you have any questions, comments or suggestions, or would like more clarification regarding the study, please do not hesitate to contact one of us at the School of Physical Education at U.VIC. If your child wishes to participate in the study and you approve, please sign the consent form below and return this complete letter to your child's teacher as soon as possible. Our intensions are to begin this study before the end of February. You have been given two copies of this letter so that you may keep one for future reference.

Your cooperation is appreciated. Thank you for your support of this project.

Sincerely,

Catherine A. Gaul (721-8392)

Dr. David Docherty (721-8385)

**EXERCISE AND THE YOUNG FEMALE
PARENTAL CONSENT FORM**

I, _____ state that I am the parent or legal guardian of _____ and do give my permission for my child to participate in the research study 'Exercise and the young female' proposed by C.A. Gaul and D. Docherty, Ph.D.

I am aware of the nature of the research and realize that there are no personal risks to my child greater than those encountered in play and sports activities. I am also aware that the researcher has explained the need for the study and that my child is free to withdraw at anytime during the study.

Signature of parent/gardian _____

Date: _____

Telephone no.: _____

Appendix C

Diet and Physical Activity Questionnaire

TO: 'Exercise and Young Females' Study Participants and Parents.

FROM: Dr. David Docherty, Professor
Catherine A. Gaul, Doctoral Student

Dear Participant and Parents,

The following questionnaire is meant to help collect some important information needed for the study in which you have volunteered to participate. Please carefully read all sections and questions and answer each as honestly and with as much detail as possible. Please note: All information will be considered confidential.

I. Biological Information

It is felt that young girls may have a different response to exercise than older girls. One of the usual ways to separate younger and older girls is to use onset of menarche to determine if a girl is sexually mature. Sexual maturity is normally defined in relation to the menstrual cycle or "period". Although this information is personal, it is important for the study, and will be treated confidentially.

A. Date of Birth _____ Age: _____

B. Have you had your first period? Yes: _____ No : _____

If 'YES':

i) When did you first start having them?

ii) How regular are they?

C. The blood lipid and cholesterol levels that we are measuring are affected by certain medications. Please indicate ANY medication you have taken within the last 6 weeks. In addition, if you are presently taking oral contraceptive (ie. the Pill), please indicate the type and how long you have been taking them (ie. MinOvral, since Sept. 1988).

D. Cigarette smoking has been linked with changes in blood cholesterol levels. It is important that we know whether or not any of our participants smoke. As with all other information, your answers will be treated confidentially.

Do you ever smoke cigarettes? Yes: _____ No : _____

If Yes: How many per day? _____ per week? _____

II. Diet Information

A. Are you on any special diet? Yes:_____ No :_____

If 'YES', Please describe this diet (ie. Vegetarian; low fat; Scarsdale...)

B. Has your diet changed recently? Yes:_____ No :_____

If 'YES', please explain WHY and HOW.

C. Do you usually eat breakfast? Yes:_____ No :_____

Please describe your typical breakfast.

Do you usually eat lunch? Yes:_____ No :_____

Please describe your typical lunch

Do you usually eat dinner? Yes:_____ No :_____

Please describe your typical dinner.

Do you regularly eat between-meal snacks?

Yes:_____ No :_____

Please describe the types of snacks you eat and when in the day you usually eat them.

- D. What sort of fluids do you normally drink each day? How much do you normally drink each day? (example: 3 glasses of 2% milk, 2 cokes, 4 glasses of water...)

III. Activity and Exercise Information

- A. Are you involved in any physical activities other than the physical education class at school? Yes:_____ No :_____

If 'YES', please describe these activities including how often you participate in them (example: swimming class 30 min, 1 time per week; Ballet class 1 hour twice a week).

B. Have you ever been on a competitive team? Yes:_____ No :_____

If 'YES', please explain the type of team and when this was (example: Softball team, 6 weeks in summer 1988)

Explain why you no longer are participating with this team.

C. Do you enjoy physical activity? Yes:_____ No :_____

Please explain your answer.

D. What sort of things do you like to do in your spare time? How much time do you usually spend doing these things? (example: Watch TV 10 hours a week; read 1 hour a day; work on computer 10 hours a week; ride bike 1 hour a day ...)

Thank you for taking the time to complete this questionnaire. The information collected from these forms will help us to complete our study. If you have any questions regarding this questionnaire or any other aspect of the study please call Kathy Gaul (721-8392) or Dr. Docherty (721-8385) at the university . Again, be assured that all information will be dealt with in a **confidential** way and will **only** be used by the investigators of this study.

Sincerely,

Kathy Gaul

PARTICIPANTS NAME: _____

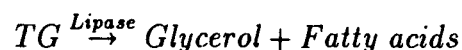
Appendix D

Assay Method for the Determination of Total Serum Triglycerides

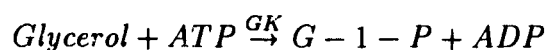
The method of the commercial kit used in this study (Diagnostic Chemicals Ltd, Charlottetown, PEI, Cat no. 210-75) has been reported to have several advantages over other conventional enzymatic methods that employ the hydrolysis of TG to release glycerol. McGowan et al. (1983) described this single reagent system as having speed, good linearity and excellent stability after reconstitution.

The principles of the assay are:

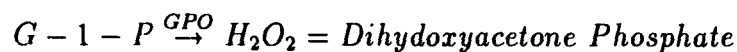
1. Serum TG are hydrolysed to glycerol and free fatty acids by Lipase:



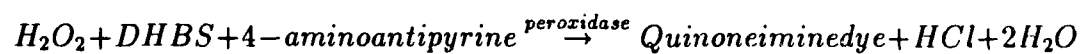
2. Glycerol is converted to glycerol-1-phosphate (G-1-P) in the presence of ATP and glycerol kinase (GK):



3. The G-1-P is oxidized to produce hydrogen peroxide (H₂O₂) by glycerol phosphate oxidase (GPO):



4. Hydrogen peroxide is condensed with DHBS and 4-aminoantipyrine in the presence of peroxidase. The resulting red colored quinoneimine dye is absorbed at 515 nm.:



The intensity of the color produced and the increase in absorbance at 515 nm is directly proportional to the concentration to TG in the sample.

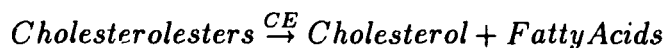
Appendix E

Assay Method for the Determination of Total Serum Cholesterol.

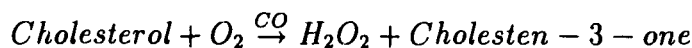
The enzymatic method used in this assay is a modification of that described by Allain et al. (1974). This assay was performed using a commercial kit (Diagnostic Chemicals Ltd, Charlottetown, PEI, Cat no. 225-26).

The principles of this assay:

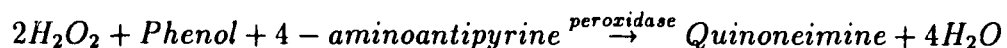
1. Hydrolysis of cholesterol esters by cholesterol esterase (CE) to form free cholesterol and fatty acids:



2. The free cholesterol is oxidized by cholesterol oxidase (CO) to hydrogen peroxide (H_2O_2) and cholesten-3-one:



3. The Hydrogen peroxide is condensed with phenol and 4-aminoantipyrine in the presence of peroxidase to yield quinoneimine:



The produced chromogen, quinoneimine, has a maximum absorbance at 505 nm. The intensity of the color produced and the increase in absorbance at 505 nm is directly proportional to the concentration of TC in the sample.

Appendix F

Video movies shown during training sessions

The following video taped movies were shown during the training sessions of the study:

Elementary School, (PreM)

Princess Bride
Grease 2
Karate Kid
Money Pit
Wizzle Wozzel Woodle Woo
Neverending Story
Romancing the Stone
Labrynth
Adventures in Baby Sitting

Junior High School, (PostM)

Princess Bride
Roxanne
Willow
Overboard
Money pit
Neverending Story
Dirty Dancing
Adventures in Baby Sitting
Can't Buy me Love

Appendix G

List of Sponsors

This study was supported by:

Canadian Fitness and Lifestyle Research Institute

University of Victoria

Be There Now Integrated Health Center

Capitol 6 Theaters

Crazy Mike's Video

Dairyland Foods

Low Cost Rentals

Mac's Cyle Center

Oak Bay Bicycles

Odeon Theaters

Picture Perfect

Russ Hay's Bicycle Shop

Rider's Cycle

Ray's Sports

Shoestrings Sports

University Bookstore

University Sports

Venice Bakeries

Appendix H

Results from Tests for Reliability

Maximal Aerobic Power

Test-retest reliability for $\dot{V}O_2$ max was determined using the two pretest $\dot{V}O_2$ max data for all subjects whose two pretest scores were considered to be maximal and not just 'peak'. Eight subjects did not reach $\dot{V}O_2$ 'max' in the first pretest and therefore, were not included in this test for reliability. A significant relationship between the pretest scores was found ($r(57)=.83, p < .001$). In addition, differences between the mean scores for each of the pretests were found to be insignificant ($t(56)=.96, p > .10$) providing evidence that the method used was reliable.

Ventilatory Threshold

As VT is a submaximal measurement, data from both pretests for all subjects were used to determine reliability of the method. A significant Pearson Product-Moment correlation coefficient ($r(65)=.67, p < .001$) and non-significant paired t-test results ($t(64)=.78, p > .10$) suggested that the results of the two pretests were not different.

Anaerobic Capacity

Since two pretest WAnT were not performed, reliability for AC was tested for using the PRETEST and TEST2 results of the combined control (C) groups. This was considered an acceptable method of determining the reliability of AC

as the C groups were not involved in any training. A strong relationship existed between the PRETEST and TEST2 AC scores for the C groups ($r(34)=.92$, $p < .001$). In addition, reliability between tests was determined from a paired t-test. No significant difference existed between the AC scores of the two test periods ($t(33)=.58$, $p > .10$).

Appendix I

Results of The Analysis of Variance for Differences Between Maturity Level and Training Level on Maximal Aerobic Power as Described in Absolute Terms, Relative to Body Weight and Relative to LBM.

	$\dot{V}O_2$ max								
	df	absolute		relative to weight		relative to LBM		relative to $H^{2.25}$	
		F	p	F	p	F	p	F	p
Maturity (M)	1,60	49.19	<.001	9.71	<.001	4.94	<.05	0.23	ns
Training (T)	1,60	1.30	ns	.53	ns	1.20	ns	0.88	ns
M x T	1,60	0.28	ns	.22	ns	0.75	ns	1.15	ns
Test	2,120	20.57	<.001	23.25	<.001	14.71	<.001	18.82	<.001
M x Test	2,120	3.12	<.05	3.98	<.05	3.30	<.01	3.21	<.05
T x Test	2,120	7.77	<.05	7.37	<.001	5.91	<.01	8.42	<.001
M x T x Test	2,120	1.11	ns	1.84	ns	2.15	ns	1.85	ns

Appendix J

**Serum 17β -Estradiol data (pmol·l⁻¹) for individual subjects
separated according to group.**

PREMENARCHEAL SUBJECTS					
Subject	Training Group		Control Group		
	PRETEST	POSTTEST	PRETEST	POSTTEST	POSTTEST
MR	80	< 50	DB	< 50	< 50
KB	< 50	< 50	TC	< 50	< 50
CC	< 50	< 50	AD	< 50	< 50
MH	< 50	< 50	RG	< 50	< 50
BM	70	70	TH	< 50	< 50
MK	< 50	< 50	JH	< 50	< 50
MAK	< 50	< 50	JL	< 50	< 50
AK	< 50	< 50	LM	100	< 50
BL	< 50	< 50	JP	< 50	< 50
SLM	< 50	< 50	RS	< 50	< 50
SM	< 50	< 50	JS	< 50	-
SS	< 50	< 50	DS	< 50	< 50

POSTMENARCHEAL SUBJECTS					
Subject	Training Group		Control Group		
	PRETEST	POSTTEST	PRETEST	POSTTEST	
KB	250	450	MB	310	170
SB	230	250	JD	120	80
YC	120	190	JE	160	1030
SC	70	240	JF	410	-
DC	180	120	SG	540	510
LC	210	190	NH	210	140
JG	500	100	WH	140	230
MG	160	150	SH	520	120
GH	340	280	AK	170	200
JJ	620	150	JL	110	-
JK	190	210	LM	720	670
MK	1030	420	SO	130	180
ANL	400	190	AP	430	-
AL	130	290	TQ	108	200
EL	780	340	ES	110	235
RL	240	340	HT	320	190
LO	120	880	TW	150	160
LAS	170	960	SW	220	140
AS	80	640	RW	430	-
LS	120	130	PW	-	210
MW	210	540			