EFFECTS OF MATURATION AND PHYSICAL ACTIVITY ON MUSCLE MASS AND STRENGTH IN PREPUBERTAL GIRLS DURING TWO-YEAR FOLLOW-UP

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ABSTRACT

Effects of Maturation and Physical Activity on Muscle Mass and Strength in Prepubertal Girls during Two-year Follow-up Minna Kontio University of Jyväskylä, Faculty of Sport and Health Sciences, Department of Health Sciences, 2005 35 pages and 1 appendix

The purpose of this study was to assess the effects of maturation and physical activity on muscle mass and strength in prepubertal girls during two-year follow-up. At baseline study subjects were 10-12 year-old and at Tanner stage I-II. They were classified into two groups according to their physical activity level over two-year period: Group 1 (consistently high physical activity, n=21) and group 2 (consistently low physical activity, n=20). Physical characteristics, muscle cross-sectional area (CSA) and isometric muscle strength were measured at baseline and at 12 and 24 months follow-up. There were no statistically significant differences in physical characteristics between the groups at baseline (p>0.05). The active group had significantly higher maximum isometric muscle strength of the elbow flexors (p=0.007) and knee extensors (p=0.007) than the inactive group during follow-up. The active group tended to have higher CSA of the lower leg muscle (p=0.073) than the inactive group during follow-up. Muscle CSA correlated with muscle strength of knee extensors among all subjects at baseline (p < 0.001), 12 months (p < 0.001) and 24 months (p < 0.001). The strongest correlation between physical characteristics and muscle variables was found for weight and muscle CSA (r=0.829, p<0.001). A significant interaction between tanner stage and physical activity on muscle CSA at baseline (p=0.05) was found. Adjustment for baseline height and weight did not change the results. In conclusion, physical activity improves muscle strength during childhood and adolescence.

Keywords: Sexual Maturation, Motor Activity, Muscles, Child

CONTENTS

1 INTRODUCTION
2 REVIEW OF THE LITERATURE
2.1 Structure and Function of Skeletal Muscle
2.1.1 Muscle Structure
2.1.2 Muscle Function
2.2 Growth and Maturation
2.2.1 Normal Growth and Maturation in Prepubertal Girls
2.2.2 Development of Muscle Mass and Strength
2.3 Physical Activity
2.3.1 Physical Activity in Prepubertal Girls
2.3.2 Effects of Physical Activity on Muscle Mass and Strength 10
3 PURPOSE OF THE STUDY 12
4 MATERIALS AND METHODS
4.1 Subjects and Background Information
4.2 Measurements of Physical Characteristics
4.3 Assessment of Physical Activity
4.4 Measurements of Muscle Cross-sectional Area and Muscle Strength14
4.5 Statistical Analysis
5 RESULTS
5.1 Physical Characteristics
5.2 Effect of Physical Activity on Muscle Mass17
5.3 Effect of Physical Activity on Muscle Strength
5.3.1 Isometric Strength of Elbow Flexors
5.3.2 Isometric Strength of Knee Extensors
5.4 Correlation between Physical Characteristics, Muscle Mass, and Muscle Strength. 20
5.5 Effects of Tanner Stage and Physical Activity on Muscle Mass and Strength 22

6 DISCUSSION	25
6.1 Key Findings	
6.1.1 Muscle Cross-sectional Area	25
6.1.2 Isometric Muscle Strength	
6.2 Other Findings	
6.2.1 Associations between Physical Characteristics and Muscle Variables	
6.2.2 Interaction of Tanner Stage and Physical Activity	
6.3 Study Limitations	
6.4 Conclusion and recommendations	29
REFERENCES	31
APPENDICES	35
Appendix 1: CALEX Physical Activity Questionnaire	

1 INTRODUCTION

J. M. Tanner started to document the normal patterns of growth and increasing strength in children in the United Kingdom about 40 years ago. That was the pioneering work and now lots of studies have been done about the normal growth and maturation in children. It is now known that growth and maturation induce increases in muscle mass and strength in children (Jones & Round 2000, 137).

Normal growth is a strong testament to the overall good health of children. More recently the effect of physical activity on growth and maturation has become a topic of interest (Rogol et al. 2002). The childhood offers the opportunity to observe muscle as it undergoes the unique process of rapid growth, development and maturation. It is not clear how physical activity or inactivity actually affects that process. In children it is possible to observe growing muscle and to identify the factors that promote its growth and increase in strength (Jones & Round 2000, 133).

Physical activity is important for performance and health during childhood, it is also a factor in the prevention of many diseases (Aarnio 2002, 15). The number of children who are not physically active during their leisure time has increased (De Knop 1998, 52). The aim of this study is to analyze the relationships between maturation, physical activity, and muscle mass and strength in prepubertal girls during two-year follow-up. This study doesn't focus on hormonal responses to muscle development in prepubertal girls.

2 REVIEW OF THE LITERATURE

2.1 Structure and Function of Skeletal Muscle

Muscle tissues are classified to: 1) skeletal muscle tissue, 2) cardiac muscle tissue, and 3) smooth muscle tissue. These three types of muscle tissues differ from each other in their microscopic anatomy, location, and control by the nervous and endocrine systems. Only the structure and function of skeletal muscle is discussed. (Tortora & Grabowski 1996, 239.)

2.1.1 Muscle Structure

Figure 1 shows the structural organisation of the skeletal muscle from macroscopic to microscopic levels. The entire skeletal muscle is typically surrounded by a fascia, epimysium. The next smaller structure is the muscle bundle called fascicle, which consists of a number of muscle fibres. The bundles of fibres are encircled by perimysial connective tissue. At the muscle fibre level, each muscle cell is surrounded by the endomysium. Muscle fibres are made up of hundreds to thousands of myofibrils. (Herzog 1999, 149, Cerny & Burton 2001, 124-125.)

The basic functional unit of skeletal muscle is the sarcomere (Figure 1). The sarcomeric units are joined together in series to form a myofibril, which are encircled by the cell membrane, the sarcolemma. Myofibrils contain even smaller myofilaments, which consist of two proteins, thin actin and thick myosin filaments. Thin filament interacts with thick filament during excitation-contraction coupling. The sarcomere consists of the basic unit between two Z lines, which give stability to the entire structure. Elastic filament, titin, helps to keep the thick filament centered between two Z lines during contraction. (Cerny & Burton 2001, 125, McArdle et al. 2001, 362, 365.)



FIGURE 1 The structure of skeletal muscle (Campbell & Reece 2002, 1081).

2.1.2 Muscle Function

Muscle cells produce contractions that move body parts. Skeletal muscle tissue is attached primarily to bones, and it produces movement of the skeleton and other parts by shortening. (Tortora & Grabowski 1996, 239, Moore & Dalley 1999, 26.) The basic mechanism of muscle contraction occurs by the sliding filament mechanism. In the relaxed state of a sarcomere, the thin filaments slide into the spaces between the thick filaments. The ends of the thin filaments derived from the successive Z lines barely begin to overlap one another, and at the same time lying adjacent to the thick filaments. In the contracted state of a sarcomere, the Z lines pull towards each other. The thin filaments have been pulled inward among the thick filaments and their ends overlap one another to a major extend. The Z lines have been pulled by the thin filament up to the ends of the thick filaments. The sliding of the thin filaments inward among the thick filaments is caused by mechanical forces generated by interaction of the cross-bridges from the thick filaments with the thin filaments. (Guyton & Hall 2000, 70.)

All muscles are composed of fast-twitch and slow-twitch muscle fibres. The fibre types are type IIA, IIB and IIC (fast-twitch fibres), and type I (slow-twitch fibres) (McArdle et al. 2001, 375). The fast-twitch fibres are adapted for rapid and powerful muscle contractions and the slow-twitch fibres are adapted for prolonged and continued muscle activity (Guyton & Hall 2000, 75).

Contraction of the muscle can be categorized into static (isometric) and dynamic (concentric and eccentric) types of actions. In isometric contraction muscle force equals the load or resistance and no movement occurs. In isometric action thin and thick filaments remain in their normal position. In concentric contraction the load is reduced and muscle force overcomes the resistance, so that the muscle will shorten. In concentric action the thin filaments are pulled closer together. In eccentric contraction the load is not reduced and the resistance overcomes muscle force, so that the muscle will lengthen. In eccentric action the thin filaments are pulled farther away from the center of the sarcomere. (Wilmore & Costill 1999, 46-47, Cerny & Burton 2001, 135-136.)

2.2 Growth and Maturation

Growth, development, and maturation are terms that can be used to describe the changes that occur with in the body. Growth can be defined as an increase in the size of the body and its parts. Development can be defined as differentiation of cells along specialized lines of function. It refers to functional changes that occur with growth. Maturation can be defined as the process of taking on adult form and becoming fully functional. (Wilmore & Costill 1999, 518.)

2.2.1 Normal Growth and Maturation in Prepubertal Girls

The somatic growth and maturation are influenced by a number of factors. The normal growth and maturation in children consist of the growth and development of bones, nerves, muscles, and organs. There are also growth-related changes in pulmonary system, cardiovascular system, and fat cells. Consequently there are changes in body size, shape, and composition. Nutrition, including energy intake, is a major determinant of growth. Under nutrition is the single most important cause of growth retardation worldwide. (Rogol et al. 2002.)

Changes in height and weight accompany growth and development. Height increases rapidly during the first two years of life. After this phase height increases at a progressively slower rate throughout childhood but just before puberty the rate increases markedly. Thereafter the rate of growth height decreases until full height is attained. The peak rate of growth in height occurs at approximately 11.5 years in girls. Change in height is assessed in terms of centimetres per year. Generally children grow 25 cm in the first year of life, half that (12 to 13 cm) in the second year and 5 to 6 cm per-year until puberty (Rogol et al. 2002). Weight increases proportionally with height. The peak rate of growth in body weight occurs at approximately 12.1 years in girls. Change in weight is assessed in terms of kilograms per year. (Wilmore & Costill 1999, 518-519.)

The state of children's maturity can be defined by stage of pubertal maturation (Wilmore & Costill 1999, 518). According to Tanner (1988, 197) children's pubertal maturation can be defined by the five scale puberty stages. Pubertal maturation stage is based on development of the breasts and pubic hair in girls (Roemmich & Rogol 1995). Children can be classified as either prepubertal (Tanner stage 1-2), peripubertal (Tanner stage 3-4), or postpubertal (post menarche) (Daly et al. 2004). The timing of the pubertal growth spurt occurs typically at the Tanner breast stage 3 in girls. The first menstrual period in girls, menarche, occurs relatively late in puberty, which is usually stage 3 or 4. The age at menarche has not decreased over the past few decades. Under nutrition is associated with later age of menarche and a moderate degree of obesity is associated with early sexual maturation. (Tanner 1988, 65-66, Rogol et al. 2002.)

2.2.2 Development of Muscle Mass and Strength

The development of muscle is a complex process and it involves several stages. The primitive structures of muscle cells are formed of cells called myotubes. In the process of the formation of new muscle fibres, each myotube becomes surrounded by an extracellural layer. As myoblasts continue to divide during maturation of the cell, some of the myoblasts get trapped between the layer and the cell membrane. These myoblasts, which are inside the basement membranes of the muscle fibres, are called satellite cells. Skeletal muscle fibres can be replaced individually by new muscle fibres derived from satellite cells. They play also a major role in the hypertrophy process. (Moore & Dalley 1999, 31, Cerny & Burton 2001, 122-123.)

Growth and maturation induce increases in muscle mass (Boisseau & Delamarche 2000). Increase in muscle mass is a result of hypertrophy of fibres (increase in size) and hyperplasia of fibres (increase in number). Increases in muscle mass with development appear to result primarily from hypertrophy, with little or no hyperplasia. Increase in fibre size results from increases in the myofibrils and myofilaments. Increase in muscle length results from increase in the number of sarcomeres and from increase in the length of existing sarcomeres. (Wilmore & Costill 1999, 523.)

Muscle strength is defined as the ability of the skeletal muscle to produce force. The muscle strength is associated with its size. A maximal contractile force is between 3 and 4 kg/cm² of muscle cross-sectional area. Strength and muscle cross-sectional areas are increasing parallel. The proportion of fast-twitch muscle fibres influences strength because fast-twitch fibres produce more force than slow-twitch fibres. The power of muscle contraction is different from muscle strength, because muscle power is measured in kilogram meters per minute. (Jackson et al. 1999, 46, Guyton & Hall 2000, 968-969.)

There is a multitude of ways in which the measurement of strength in children can be assessed. For example isokinetic dynamometry, hand-held dynamometry, field tests, and standard weights equipment can be used to assess muscle function in children (Jones & Stratton 2000). According to Jones and Round (2000, 136) measurement of isometric force of major muscle groups, such as the knee extensors (quadriceps) and the forearm flexors (mainly biceps), represents the best option for physiological assessment. Also many longitudinal studies have measured isometric grip strength and it has been found that girls increase grip strength in a linear fashion throughout childhood (Roemmich & Rogol 1995).

As children's size increases, so does their functional capacities as strength. (Wilmore & Costill 1999, 518). It is generally agreed that during childhood there is a steady increase in strength with little difference seen between boys and girls until puberty (Jones & Round 2000, 137). Different physical factors such as variations in height and gains in body weight have been associated with changes in muscle strength during growth and maturation (Ramos et al. 1998). The development of strength relates to other parameters of growth during prepubertal period when the sex steroids have no role to play. For girls a curvilinear relationship between strength of the elbow flexors and height (r=0.69) has been found. (Jones & Round 2000, 137-138.) The muscle mass steadily increases with age and along with the weight gain. (Wilmore & Costill 1999, 523).

Neu et al. (2002) analyzed the relationship between cross-sectional area of forearm muscles and maximal isometric grip force with age and pubertal stage. Both muscle cross-sectional area and grip force were higher in prepubertal boys than in girls. The gender differences decreased until pubertal stage 3. They found out that forearm muscle growth takes a gender-specific course during puberty. Jones and Round (2000, 137) have measured

isometric strength in post-menarchial girls in the mid 1990s and they found no evidence of a loss of strength after menarche. They note that there has also been found that girls seem to decrease in strength after menarche.

The development of strength is influenced by myelination and maturation of the nervous system. High level of strength is impossible if the child has not reached neural maturity. (Wilmore & Costill 1999, 524-525, 527.) It is also good to remember that the muscle system is an important predictor for bone development in children. Schoenau et al. (2000) have studied the relationship between muscle and bone development before and during puberty. They found that there was a strong correlation between muscle area and cortical area of the radius in children (r=0.88).

2.3 Physical Activity

Physical activity is defined as bodily movement that is produced by the contraction of skeletal muscle. Exercise is defined as planned, structured, and repetitive bodily movement done to improve or maintain one or more components of physical fitness. Physical fitness is defined as a set of attributes that relates to the ability to perform physical activity. (American College of Sports Medicine 2000, 4, Kesaniemi et al. 2001.)

2.3.1 Physical Activity in Prepubertal Girls

The promotion of public health is a reason for paying attention to physical activity among children. Physical activity is important for performance and health during childhood. The beneficial affects of physical activity on health have been well studied. (Telama 1998, 64.) Physical activity produces a number of major health benefits and it is a factor in the prevention of many diseases. Regular physical activity have been associated with a reduction in all-cause mortality, cardiovascular disease, coronary heart disease, and the incidence of obesity, type 2 diabetes mellitus, colon cancer, hypertension, low back pain, depression, and osteoporosis. Many of the risk factors for coronary artery disease,

9

hypertension, non-insulin-dependent diabetes and osteoporosis appear to develop in childhood. Physical activity has favourable metabolic effects on the development of fat tissue, skeleton, tendons, ligaments, and cartilage. (Kesaniemi et al. 2001, Aarnio 2002, 15.)

The two principal categories of physical activity are occupational and leisure-time. The dose of physical activity is described by the characteristics of frequency, duration, intensity and type of activity. Frequency is described as the number of activity sessions per time period and duration refers to the number of minutes of activity in each session. Intensity describes the effort associated with the physical activity. (Howley 2001.) Physical activity variables among children has been measured in many ways: from habitual physical tasks to sports club activities, and from direct oxygen consumption measurements to questionnaires (Aarnio 2002, 17). For example Telama (1998, 66) used variables such as leisure-time activities, physical activities, and sport participation which were measured with a questionnaire.

Grund et al. (2000) assessed different aspects of physical activity in order to develop a basis for sport programmes for overweight and obese prepubertal children. In their study physical activity level was calculated from total energy expenditure and resting energy expenditure. Muscle strength was inversely associated with fat mass in 7.6-11.4 year-old children. They found out that increased fitness and reduced physical inactivity may prevent children from being overweight.

According to De Knop (1998, 52) children are either training intensively several times a week or they are not physically active at all during their leisure time. The variety between children concerning physical activity is increased and therefore the number of children who are physically inactive has increased. De Knop (1998, 56) studied the trends in youth sport in the international project "Children, Youth and Sport", which included youth sport data also from Finland. De Knop (1998, 59) found that the average age of starting participation in sport has decreased. Children start to participate in sports at the age of seven or eight or even younger but the number of dropouts is increasing with increasing age, especially among girls. The dropout problem among girls was reported from 20 countries in the study.

Sleap and Tolfrey (2001) investigated whether a sample of 9-12 year-old children fulfilled existing United States and United Kingdom physical activity recommendations for health. They studied habitual physical activity levels of 79 pre- and early pubertal children. They found out that British children appear to engage in sufficient physical activity to meet the U. S. and UK minimum daily recommendations.

2.3.2 Effects of Physical Activity on Muscle Mass and Strength

Muscle development is specific and only the muscle fibres that are engaged in the activity can increase in strength (Jackson et al. 1999, 194). Physical capacities such as muscle strength, endurance and power are dependent on body or muscle mass. To determine whether age-related changes are due to an increase in mass or to other maturational factors, variables are normalized in some manner. For example, strength might be expressed per lean muscle mass or cross-sectional area of the involved muscles. (Cerny & Burton 2001, 250.) The data suggest that there is an increase in size without a change in fibre numbers as the muscles grow in size and strength. A comparison of quadriceps cross-sectional area and muscle fibre areas showed that muscle fibre area was the main factor determining cross-sectional area of the whole muscle. (Jones & Round 2000, 134.)

Changes in the muscles with growth and maturation can greatly affect physical activity and exercise performance. Some of these changes are related to the muscle metabolic capability. This metabolic capability of the developing muscles shows higher oxidative enzyme activities in the children compared to those for adults. (Cerny & Burton 2001, 249-250.) The physiological responses to activity are dependent on the energy requirements of the activity. Performing physical activity requires energy. The source of energy for muscle contraction is adenosine triphosphate (ATP). (Cerny & Burton 2001, 1, 23.) Physical activity has the most profound effect on human energy expenditure (McArdle et al 2001, 192).

During growth and maturation, the study of very brief high-intensity exercise has not received the same attention as aerobic function. In anaerobic sports events the children's performance is distinctly lower than that of adults. This reflects children's lesser ability to

generate mechanical energy from chemical energy sources during short-term intensive activity. Paediatric exercise scientists have concentrated on measuring short-term muscle power by short-term cycling power tests, vertical jump tests and running tests. The results support the hypothesis that the difference observed between children and adolescents during short-term muscle power testing is related to neuromuscular factors, hormonal factors and improved motor coordination. (Van Praagh & Dore 2002.)

One approach to determining the effects of physical activity on development is to examine the effects of physical training on muscle strength (Cerny & Burton 2001, 254). Faigenbaum et al. (2002) compared the effects of 1 and 2 days per week of strength training on upper body strength, lower body strength, and motor performance ability in children. 21 girls and 34 boys between the ages of 7.1 and 12.3 years volunteered to participate in the study. Participants who strength trained either once per week (n=22) or twice per week (n=20) for 8 weeks and children (n=13) who did not strength train served as age-matched controls. 1RM strength on the chest press and leg press, grip strength, long jump, vertical jump, and flexibility were assessed at baseline and post-training. Only participants who strength trained twice per week made significantly greater gains in 1RM chest press strength compared to the control group (p<0.05). On average, participants who strength trained once per week achieved 67% of the 1RM strength gains. The findings support the concept that muscular strength can be improved during childhood.

Eliakim et al. (2001a) studied the effect of training in pre- and early pubertal nonobese American girls. Forty girls (mean age 9.1 year) enrolled in a 5 day/week summer school program for 5 weeks and were randomized to a control (n=20) or a training groups (n=20; 1.5 h/day, endurance-type exercise). Total energy expenditure was measured by a double labeled water and thigh muscle volume magnetic resonance imaging. Total energy expenditure was significantly greater (p<0.02) in the training girls and the training also increased thigh muscle volume (p<0.005). Thigh muscle volume per weight was much lower in adolescent compared with prepubertal girls (p<0.001). They found that muscle function was quite responsive to brief training in prepubertal girls and therefore increase in muscle mass may be depressed in nonobese American girls as they mature.

3 PURPOSE OF THE STUDY

The normal growth and maturation induce increases in muscle mass and strength in children (Boisseau & Delamarche 2000). It is known that during childhood there is a steady increase in strength with little difference seen between boys and girls until puberty (Jones & Round 2000, 137). Different physical factors such as variations in height and gains in body weight have been associated with changes in muscle strength (Ramos et al. 1998). It is also known that muscular strength can be improved during childhood by physical training (Faigenbaum et al. 2002). The effect of physical activity on muscle mass and strength is a less studied area.

The purpose of this study is to evaluate the relationships between maturation, physical activity, and muscle mass and strength in prepubertal girls during two-year follow-up. This study doesn't focus on hormonal responses to muscle development. The aim of the study is to find out:

What is the effect of physical activity on muscle development?

- 1) What is the effect of physical activity on muscle mass?
- 2) What is the effect of physical activity on muscle strength?

4 MATERIALS AND METHODS

4.1 Subjects and Background Information

Study subjects were girls who enrolled in the CALEX-study (Effect of calcium, vitamin D supplementation, and physical activity on bone mass accrual in prepubertal girls). Subjects were healthy girls aged 10-12 years at the beginning of the study and they lived in the city of Jyväskylä and its surrounding region in Finland. Subjects were at Tanner stage I-II of pubertal maturation and they had no medications or diseases that could affect bone metabolism. Subjects were assessed at baseline and after 6, 12, 18 and 24 months follow-up. Informed consent was obtained from all participants and their parents.

There were 258 girls who were eligible for the CALEX-study. In this study, it is only consists a sub-group of girls who were physically active (n=21) and inactive (n=20) with maturational stage I-II (prepubertal) at baseline. Subject had to be persistently active (active at baseline and 12 and 24 months follow-up) or persistently inactive (inactive at baseline and 12 and 24 months follow-up) to be included in the study.

4.2 Measurements of Physical Characteristics

Physical characteristics measurements included age, body height, body weight, body mass index, and Tanner stage. These were used to assess physical development of the girls. Date of birth was used to calculated age in years. Height (cm) was measured using stadiometer to the nearest 0.1 cm. Weight (kg) was measured using scale to the nearest 0.1 kg. Height and weight were determined with subjects wearing only light clothing and no shoes. Height and weight were used to calculate body mass index (BMI) which is expressed as kg/m² (weight (kg)/height² (m)). Sexual maturation was assessed together by the girl and the study nurse by the standard five scales Tanner puberty stages (Tanner 1988, 197). These were measured at baseline and 6, 12, 18 and 24 months follow-up.

4.3 Assessment of Physical Activity

Current physical activity was assessed by self-reported questionnaire that detailed the frequency, duration, and type of physical activity (Appendix 1). Subjects engaged in physical education lessons at school were not included in the questionnaire. The following question was used to evaluate the kisure-time physical activity (Appendix 1):

Outside school classes: Mark, how many hours a week you usually exercise in your free time so much that you get out of breath and sweat

1. None

2.	About 1/2 hour	
3.	About 1 hour	
4.	About 2-3 hours	
5.	About 4-6 hours	
6.	7 hours or more	

Subjects were classified as physically active if they exercised at least 4 hours a week in their leisure time. This means that girls had to answer to this question alternative 5 or 6 at baseline and 12 and 24 months follow-up. Subjects were classified as physically inactive if they exercised at most 1 hour a week in their free time. This means that girls had to answer to this question alternative 1, 2, or 3 at baseline and 12 and 24 months follow-up.

4.4 Measurements of Muscle Cross-sectional Area and Muscle Strength

Muscle cross-sectional area (CSA) of the lower leg muscles (mm²) was measured by peripheral quantitative computed tomography (pQCT) (XCT 2000, Stratec GmbH, Germany). The pQCT measurement was performed at the left tibia shaft, 60 % upwards from the medial malleolus of the length of the lower leg. The length of the lower leg was the distance between the tuberositas tibia and the medial malleolus which was measured in a sitting position with a knee angle of 90 degrees. Muscle CSA was measured at baseline and 12 and 24 months follow-up.

Maximum isometric muscle strength (N) was measured in a sitting position with a dynamometer of isometric muscle strength (Metitur, Finland). Isometric strength test were maximum force (N) of the left knee extensors (quadriceps) and maximum force (N) of the left elbow flexors (mainly biceps). Maximum isometric strength was measured three times and the highest values were used in the analysis. Muscle strength was measured at baseline and at 6, 12, 18 and 24 months follow-up.

4.5 Statistical Analysis

Descriptive statistics (means and standard deviations) for age, height, weight, body mass index, muscle cross-sectional area, and isometric muscle strength tests were computed in both physical activity groups. Independent-samples \pm test was used to detect differences between study groups in baseline variables (age, height, weight and body mass index). Median test was used to detect differences between study groups in Tanner stage values at baseline. Analysis was carried out using the SPSS 11.5 for student version. Statistical significance was taken at the *p*-value less than 0.05 level in all of the analyses.

Analysis of variance (ANOVA) for repeated measures, with repeated contrast, was used to determine the effect of physical activity on muscle cross-sectional area and isometric muscle strength with time serving as the within-subjects factor and level of physical activity as the between-subjects factor. Pearson's correlation coefficients were used to assess correlation between physical characteristics, muscle mass, and muscle strength. Two-way analysis of variance (2-ANOVA) was used to determine the effects of tanner stage and physical activity on muscle cross-sectional area and isometric muscle strength.

5 RESULTS

5.1 Physical Characteristics

The study subjects were classified into two groups according to their physical activity level: Group 1 (physically active subjects) and group 2 (physically inactive subjects). There were no statistically significant differences between the groups at the beginning of the study. The physical characteristics of the two groups are presented in Table 1. There were no significant differences in baseline age, height, weight, and body mass index (BMI) between the groups (p>0.05). Figure 2 shows the tanner stages of active and inactive subjects at baseline. There was no significant difference in baseline tanner stage between the groups (x^2 =0.344, p=0.558).

	Group 1 (n=21)	Group 2 (n=20)	
Variable	Mean (SD)	Mean (SD)	<i>p</i> value (t-test)
Age (years)	11.03 (0.73)	10.87 (0.73)	<i>p</i> >0.05
Height (cm)	143.97 (8.67)	141.23 (7.23)	<i>p</i> >0.05
Weight (kg)	36.28 (7.18)	33.28 (6.25)	<i>p</i> >0.05
BMI (kg/m²)	17.38 (2.08)	16.59 (2.18)	<i>p</i> >0.05

TABLE 1 Physical characteristics of active (group 1) and inactive (group 2) subjects at baseline.



FIGURE 2 Tanner stages of active (group 1) and inactive (group 2) subjects at baseline.

5.2 Effect of Physical Activity on Muscle Mass

The values for muscle CSA of the lower leg muscles are presented in Figure 3. There was a trend that physically active girls had greater muscle CSA at all the measured but there were no statistically significant differences between the groups (p=0.056 at baseline, p=0.167 at 12 months and p=0.053 at 24 months).



FIGURE 3 Muscle cross-sectional area of active (group 1) and inactive (group 2) subjects during two-year follow-up.

Figure 3 also shows the results of the repeated measurements analysis for muscle CSA of the lower leg muscles in active and inactive subjects during two-year follow-up. There was no statistically significant interaction (F=0.823, p=0.409) between time (within-subjects) and physical activity level (between-subjects). A significant increase in muscle CSA with time was found in both groups (F=103.4, p<0.001). There was a tendency to difference in the main effect of the physical activity level (F=4.403, p=0.073).

5.3 Effect of Physical Activity on Muscle Strength

5.3.1 Isometric Strength of Elbow Flexors

The values for the maximum isometric muscle strength of the elbow flexors are presented in Figure 4. There was no significant difference in isometric muscle strength between the groups at the beginning of the study p=0.063). There were significant differences in isometric muscle strength between the groups at the 12 months (p=0.048) and 24 months (p=0.002) follow-up.



FIGURE 4 Maximum isometric muscle strength of elbow flexors of active (group 1) and inactive (group 2) subjects during two-year follow-up.

Figure 4 also shows the results of the repeated measurements analysis for maximum isometric muscle strength of the elbow flexors in active and inactive subjects during the two-year follow-up. There was a marginally significant interaction (F=3.086, p=0.051) between time (within-subjects) and physical activity level (between-subjects). Isometric muscle strength with time was found to be significantly increased in both groups (F=39.73, p<0.001), and also the main effect of the activity level was statistically significant (F=8.064, p=0.007).

5.3.2 Isometric Strength of Knee Extensors

The values for the maximum isometric muscle strength of the knee extensors are presented in Figure 5. There was no significant difference in isometric muscle strength between the groups at the beginning of the study p=0.103). There were significant differences in isometric muscle strength between the groups at the 12 months follow-up (p=0.016) and 24 months follow-up (p=0.002).



FIGURE 5 Maximum isometric muscle strength of knee extensors of active (group 1) and inactive (group 2) subjects during two-year follow-up.

Figure 5 also shows the results of the repeated measurements analysis for maximum isometric muscle strength of the knee extensors in active and inactive subjects during the two-year follow-up. There was a statistically significant interaction (F=6.040, p=0.006) between time (within-subjects) and the activity level (between-subjects). Isometric muscle strength with time was found to be significantly increased in both groups (F=63.73, p<0.001). The active group increased more by contrast to the inactive group (F=8.043, p=0.007 main effect of the activity level).

5.4 Correlation between Physical Characteristics, Muscle Mass, and Muscle Strength

Pearson's correlation coefficients between physical characteristics and muscle CSA at baseline are presented in Table 2. Correlation coefficients are presented for all subjects, and for active and inactive subjects separately. Except for age, physical characteristics were positively associated with muscle cross-sectional area.

	Muscle cross-sectional area (mm ²)		
	All subject	Group 1	Group 2
	(n=41)	(n=21)	(n=20)
	Correlation (<i>p</i> value)	Correlation (<i>p</i> value)	Correlation (p value)
Age (years)	0.253	0.188	0.269
	(0.111)	(0.415)	(0.251)
Height (cm)	0.541**	0.548*	0.506*
	(<0.001)	(0.010)	(0.023)
Weight (kg)	0.829**	0.805**	0.854**
	(<0.001)	(<0.001)	(<0.001)
BMI (kg/m²)	0.761**	0.702**	0.798**
	(<0.001)	(<0.001)	(<0.001)

TABLE 2 Correlation between physical characteristics and muscle CSA of all subjects, active (group 1) and inactive (group 2) subjects at baseline.

* Correlation is significant at the 0.05 level

** Correlation is significant at the 0.01 level

Pearson's correlation coefficients between physical characteristics and maximum isometric muscle strength of elbow flexors and knee extensors at baseline are presented in Table 3. Correlation coefficients are presented for all subjects, and for active and inactive subjects separately. There were positive associations between age, weight, and body mass index (BMI) and isometric muscle strength. There was a weak association between height and isometric muscle strength.

	Max. isometric strength of elbow flexors (N)		Max. isom e	netric streng extensors (N	th of knee ()	
	All subject (n=41)	Group 1 (n=21)	Group 2 (n=20)	All subject (n=40) ¹	Group 1 (n=20) ¹	Group 2 (n=20)
	Corr	elation (p va	alue)	Corr	elation (p v	alue)
Age (years)	0.396*	0.487*	0.258	0.404**	0.497*	0.296
	(0.010)	(0.025)	(0.271)	(0.010)	(0.026)	(0.205)
Height (cm)	0.159	0.222	-0.034	0.298	0.191	0.368
	(0.322)	(0.335)	(0.888)	(0.062)	(0.421)	(0.111)
Weight (kg)	0.350*	0.291	0.326	0.470**	0.320	0.572**
	(0.025)	(0.201)	(0.161)	(0.002)	(0.169)	(0.008)
BMI	0.375*	0.218	0.480*	0.471**	0.337	0.535*
(kg/m²)	(0.016)	(0.342)	(0.032)	(0.002)	(0.146)	(0.015)

TABLE 3 Correlation between physical characteristics and isometric muscle strength of all subjects, active (group 1) and inactive (group 2) subjects at baseline.

¹ n=40/n=20 because one value is missing

* Correlation is significant at the 0.05 level

** Correlation is significant at the 0.01 level

Pearson's correlation coefficients between muscle CSA of the lower leg muscles and maximum isometric muscle strength of the knee extensors are presented in Table 4. Correlation coefficients are presented for all subjects, active and inactive subjects at baseline and 12 and 24 months follow-up. When data were presented separately by time, was found that muscle CSA correlated with isometric muscle strength only in the inactive subjects. No significant association between muscle CSA and isometric muscle strength in the active subjects was found.

	All subjects (n=40)	Group 1 (n=20)	Group 2 (n=20)
	Correlation (<i>p</i> value)	Correlation (<i>p</i> value)	Correlation (p value)
Baseline	0.515**	0.375	0.561*
	(0.001)	(0.104)	(0.010)
12 months	0.499**	0.303	0.636**
	(0.001)	(0.194)	(0.003)
24 months	0.515**	0.310	0.619**
	(0.001)	(0.183)	(0.004)

TABLE 4 Correlation between muscle CSA and isometric muscle strength of knee extensors of all subjects, active (group 1) and inactive (group 2) subjects at baseline and 12 and 24 months follow-up.

* Correlation is significant at the 0.05 level

** Correlation is significant at the 0.01 level

5.5 Effects of Tanner Stage and Physical Activity on Muscle Mass and Strength

Figure 6 shows the results of the two-way ANOVA for muscle CSA of the lower leg muscles in active and inactive subjects at different pubertal maturation stages (Tanner stage I and II) at the baseline. Significant interaction between tanner stage and physical activity was found to the muscle CSA (F=4.095, p=0.05). Significant effect of tanner stage was found within inactive subjects (F=12.66, p=0.001) but not within active subjects (F=0.879, p=0.354) for the muscle CSA. Significant effect of physical activity was found in subjects with tanner stage I (F=7.483, p=0.010) but not in those with tanner stage II (F=0.436, p=0.513).



FIGURE 6 Muscle CSA of active (group 1) and inactive (group 2) subjects at different pubertal maturation stages (Tanner stage I and II) at baseline.

Figure 7 shows the results of the two-way ANOVA for maximum isometric muscle strength of elbow flexors in active and inactive subjects at different pubertal maturation stages (Tanner stage I and II) at baseline. No statistically significant interaction between tanner stage and physical activity was found (F=0.014, p=0.905), nor was the main effect of tanner stage (F=2.127, p=0.153) or physical activity (F=2.420, p=0.128) statistically significant.



FIGURE 7 Maximum isometric muscle strength of elbow flexors of active (group 1) and inactive (group 2) subjects at different pubertal maturation stages (Tanner stage I and II) at baseline.

Figure 8 shows the results of the two-way ANOVA for maximum isometric muscle strength of knee extensors in active and inactive subjects at different pubertal maturation stages (Tanner stage I and II) at baseline. No statistically significant interaction between tanner stage and physical activity was found (F=0.426, p=0.518). Tanner stage has significant effect on the maximum strength (F=4.546, p=0.040) but no effect of physical activity was found (F=1.196, p=0.281).



FIGURE 8 Maximum isometric muscle strength of knee extensors of active (group 1) and inactive (group 2) subjects at different pubertal maturation stages (Tanner stage I and II) at the baseline.

6 DISCUSSION

6.1 Key Findings

The purpose of this study was to assess the relationships between maturation, physical activity, and muscle mass and strength in prepubertal girls during a two-year follow-up. This study didn't focus on hormonal responses to muscle development in prepubertal girls. The aim of the study was to find out the effect of physical activity on muscle development. No statistically significant differences in baseline variables between the physically active and inactive subjects were found. The main findings of this study are:

- Despite a directional tendency no statistically significant difference was observed in muscle mass between the physically active and inactive girls during the two-year follow-up (Figure 3).
- 2. A statistically significant difference was observed in isometric muscle strength between the physically active and inactive girls during the two-year follow-up (Figure 4 and Figure 5).

6.1.1 Muscle Cross-sectional Area

The first aim of the study was to find out the effect of physical activity on muscle mass. Analysis of variance for repeated measures was used to assess the differences in muscle CSA between the physically activity groups during the two-year follow-up. No statistically significant interaction or effect of the physical activity level were found. A significant increase in muscle mass with age was observed in both activity groups.

There was a difference in muscle CSA between the groups during the two-year follow-up but this was not statistically significant (p=0.073). One of the explanations for these results can be the number of subjects used, there were only 21 physically active and 20 inactive subjects. Many factors, such as the chronological age, can influence muscle mass. According to Boisseau and Delamarche (2000) there is an increase in muscle mass with

age. The same trend was seen in these results and an increase in muscle mass with age was observed in all subjects despite the activity levels.

The performance will depend on the size and strength of an individual muscle but also on their coordinated activation (Jones & Round 2000, 136). The ability to produce tension proportional to muscle CSA is lower in children than in adults. Resistance training studies in children have determined that regular stimulus in motor unit enhances the ability to generate muscle tension through improved neural adaptation rather than an increase in muscle CSA. In situations where longitudinal assessment is required neural adaptations may be the cause of change rather than variation in muscle CSA. (Jones & Stratton 2000.) According to Van Praagh and Doré (2002) there are cases where increases in muscle force occur without any increase in muscle CSA. This can be explained by an increases in the capacity to activate more motor units.

6.1.2 Isometric Muscle Strength

The second aim of the study was to find out the effect of physical activity on muscle strength. Analysis of variance for repeated measures was used to assess differences in isometric muscle strength of elbow flexors between the activity groups during the two-year follow-up. No statistically significant interaction was found but a statistically significant effect of activity level was found. Significant increase in muscle strength with age was observed in both activity groups. Analysis of variance for repeated measures was also used to assess the differences in isometric muscle strength of knee extensors between the activity groups during the two-year follow-up. Significant interaction and effect of the physical activity level were found. Significant increase in muscle strength with age was also observed in both activity groups.

It is known that during childhood there is a steady increase in strength until puberty (Jones & Round 2000, 137). The same trend was seen in this study and the increase in muscle strength with age was observed in all subjects despite the activity level. These results are in accordance with previous findings in studies of children. According to previous studies

(Eliakim et al. 2001a, Faigenbaum et al. 2002) muscle strength can also be improved by physical training or exercise.

The method of testing muscle function in children should reflect the purpose of the assessment as closely as possible. Isometric muscle test measures the tension developed at a specific point in the range of movement under static conditions. A limitation of isometric muscle test is that the optimal angles for tension development remain undetermined in children. Isometric tests offer a validated tool for measuring muscle strength as they measure the ability of the muscle to exert maximal tension. It appears that isometric tests are reliable when the same experimenter repeatedly tests the same population. (Jones & Stratton 2000.)

6.2 Other Findings

6.2.1 Associations between Physical Characteristics and Muscle Variables

Physical characteristics, except age, were positively associated with muscle CSA. The strongest correlation was found between weight and muscle CSA. There were also positive associations between isometric muscle strength and age, weight and body mass index. There was only a weak association between height and isometric muscle strength. There was a significant positive association between muscle CSA and isometric muscle strength. There inactive subjects compared to the active subjects.

Many studies have been done on the normal growth and maturation in children. Therefore it is known that growth and maturation induce increases in muscle mass and strength in children. Different physical factors have been associated with changes in muscle strength during growth and maturation. According to Jones & Round (2000) strength increases in proportion to body size. Compared to other studies an association has been found between strength of the elbow flexors and height (r=0.69). In this study the same association was weak (r=0.159) but associations between the other physical characteristics and strength of elbow flexors were higher. These findings support the idea that physical factors, such as age, height, weight and body mass index, have associations with muscle mass and strength.

6.2.2 Interaction of Tanner Stage and Physical Activity

Two-way ANOVA was used to detect possible interactions between tanner stage and physical activity with relation to muscle mass and strength Significant interaction between tanner stage and physical activity on muscle CSA was found. Significant effect of tanner stage within inactive subjects and significant effect of physical activity within tanner stage I were found. No statistically significant interaction between tanner stage and physical activity on isometric muscle strength of elbow flexors or knee extensors were found. Significant effect of tanner stage on isometric muscle strength of knee extensors was found. Similar findings were obtained for all variables after analysis of covariance (ANCOVA) using physical characteristics as the covariates.

The Tanner pubertal maturation stages were used to assess girls' degree of biologic development. According to Roemmich and Rogol (1995) muscle mass and strength are more related to the level of maturation than to the chronological age. This study found interaction between tanner stage and physical activity on muscle mass but not on muscle strength. The interactions among these variables on muscle mass are more important than their simple effects. These findings support the idea that both maturation and physical activity effects on muscle mass and strength.

6.3 Study Limitations

It is important to understand the definition of physical activity when studying physical activity in children. The researcher has to decide how to define physical activity and how to measure it. In this study the leisure-time physical activity was assessed by a self-reported questionnaire. Standardised technique to measure physical activity in children does not exist. Each method of assessing physical activity has strengths and limitations for its use in studies of physical activity and health-related outcomes (Lamonte & Ainsworth 2001).

Variability in childhood development must be taken into account when evaluating physical activity responses. It is difficult to separate the normal growth of muscle from the effects of physical activity on muscle development. Different physical factors, such as age, height and weight, have been associated with changes in muscle strength during growth (Ramos et al. 1998). The changes in muscles with growth and maturation can also greatly affect physical activity and exercise performance.

More recently the effect of physical activity on growth and maturation has become a topic of interest. Physical activity is important for performance and health during childhood. Moderate physical activity is associated with cardiovascular benefits and favourable changes in body composition in children. It is a factor in the prevention of many diseases. On the other hand excessive physical activity or high-intensity training during childhood may negatively affect growth and development (Rogol et al. 2002).

It is also good to remember that hormonal factors have an effect on muscle growth and development. The hormonal regulation of the growth and the changes in body composition depend on the release of the gonadotropins, leptin, the sex-steroids, thyroid hormone, and growth hormone (Ramos et al. 1998, Rogol et al. 2002). Studies of animal models have shown that Igf-I acts directly on muscle growth. Insulin-like growth factor I (Igf-I) is a peptide that mediates the effects of growth hormone in humans and animals. (Beaune et al. 1997.)

Literature was searched using Medline and the Jyväskylä University Library. The key words in use were growth, development, maturation, physical activity, exercise, muscle, prepubertal, and children. Probably all of the studies were not found because the literature was not searched systematically.

6.4 Conclusion and recommendations

The main results of this study were the significant differences in isometric muscle strength between the activity groups during the follow-up. The same trend was seen in muscle CSA but the difference was not significant. As previous study shows (Jones & Round 2000, 137)

a significant increase in muscle mass and strength with age was observed in all subjects. These findings support the idea that physical activity improves muscle strength during childhood and adolescence.

To promotion of public health is a reason for taking physical activity into consideration in children. The world wide problem is now that children are becoming less physically active and they are getting overweight. For example according to Eliakim et al. (2001b) American girls are now becoming less physically active in early puberty. Obesity is increasing in children and a significant relationship between activity levels and body fat in children has been identified (Rowlands et al 2000). The benefits of regular physical activity in children are well documented but many children are not engaged enough in physical activity to be associated with health benefits (Crocker et al. 2000). Optimal level of physical activity during childhood may reduce obesity and prevent osteoporosis and cardiovascular disease in adulthood (Eliakim et al. 2001b).

Many studies have been done about the development of muscle mass and strength in children. The effects of physical activity on muscle mass and strength in children is less studied area. There exist no other study compared to this one and these kind of results have not been previously reported. All that makes these results more remarkable. Regular physical activity can improve muscle strength but physical activity is only one of the factors linked with health.

The results of this study suggest that physical activity has an effect on muscle development. Most importantly, this study indicates that regular physical activity improves muscle strength. More longitudinal studies are needed, using better methods to measure physical activity, to determine the effects of maturation and physical activity on muscle development in both sexes and different ages. More longitudinal studies are also needed to determine whether physical activity results in a statistically significant increase in muscle CSA. In the future, more studies about the effects of physical activity on muscle mass and strength in children are needed.

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CALEX Physical Activity Questionnaire				
Study:	Date:			
Participant Initials:	_ Randomization ID:			
Group: Visit:	Screening ID:			

- 1. **Outside school classes**: Mark, how often you usually exercise in your free time so much that you get out of breath and sweat.
 - 1. Less than once a week
 - 2. Once a week
 - 3. 2-3 times a week _____
 - 4. 4-6 times a week _____
 - 5. Daily _____
- 2. **Outside school classes**: Mark, how many hours a week you usually exercise in your free time so much that you get out of breath and sweat
 - 1. None
 - 2. About 1/2 hour
 - 3. About 1 hour
 - 4. About 2-3 hours
 - 5. About 4-6 hours
 - 6. 7 hours or more
- 3. Are you more physically active in school or during your free time?

School ____ Free time ____

Appendix 1/ 2

CALEX	Physical	Activity	Questionnaire
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Study:		Date:	
Participant In	nitials:	_ Randomization ID:	
Group:	Visit:	Screening ID:	

4. What kind of physical activities do you usually go in for in your free time? Write three of your most favorite physical activities?

In summer	1)	2)	3)
T	1\	2	
In winter	1)		3)

5. For how long on average, on any single occasion do you participate in this kind of exercise?

In summer:		
1)	2)	3)
1. under 30 minutes _	 1. under 30 minutes	 1. under 30 minutes
2. 30-60 minutes _	 2. 30-60 minutes	 2. 30-60 minutes
3. 1-2 hours _	 3. 1-2 hours	 3. 1-2 hours
4. over 2 hours _	 4. over 2 hours	 4. over 2 hours

In winter:		
1)	2)	3)
1. under 30 minutes	1. under 30 minutes	1. under 30 minutes
2. 30-60 minutes	2. 30-60 minutes	2. 30-60 minutes
3. 1-2 hours	3. 1-2 hours	3. 1-2 hours
4. over 2 hours	4. over 2 hours	4. over 2 hours

CALEX Ph	ysical Activ	vity Quest	ionnaire
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Study:	Date:
Participant Initials:	Randomization ID:
Group: Visit:	Screening ID:

- 6. Are you a member of a sport club?
 - _____1. No
 - _____ 2. Yes, and I am training in a sports club
 - _____ 3. Yes, but I don't participate
- 7. About how many times per week do you usually do heavy household chores, such as scrubbing floors, vacuuming, sweeping, yard work, gardening, or snow shoveling? And for about how many hours per time?

times per week _____ hours per time _____

8. During an average 24-hour day, about how many hours do you usually spend **sleeping** and **lying down with your feet up**? (Be sure to include time sleeping at night of trying to sleep, resting or stretched out on the sofa watching TV etc.) Also about how many hours do you usually spend **sitting upright**? (Be sure to include time sitting at the table eating, driving or riding in a car or bus, sitting watching TV etc.)

Circle the best answer

During vacations and weekends Hours/Day

hours lying down:	6	7	8	9	10	11	12	13	14
hours sitting:	2	3	4	5	6	7	8	9	10

During School time			Ho	ours/D	Day				
hours lying down:	6	7	8	9	10	11	12	13	14
hours sitting:	2	3	4	5	6	7	8	9	10

Appendix 1/ 4

CALEX Physical Activity Questionnaire				
Study:	Date:			
Participant Initials:	Randomization ID:			
Group: Visit:	Screening ID:			

Attitude towards sports and physical activities

- 9. Do you think that you will go in for sports or other physical activies when you are 20 years old?
 - ____1 Definitely yes
 - _____ 2 Probably yes
 - _____ 3 Probably no
 - _____4 Definitely no

10. How good are you at sports, compared to others the same age as you?

- _____1 Among the best
- _____ 2 Good

_____ 3 Average

- _____ 4 Below average
- 11. What do you think of your physical education lessons at school?
 - ____1 Like them very much
 - ____2 Like them
 - _____ 3 Neither like nor dislike them
 - _____4 Dislike them
 - _____5 Dislike them very much
 - 6 Do not attend them

Appendix 1/ 5

CALEX Physical Activity Questionnaire				
Study:	Date:			
Participant Initials:	Randomization ID:			
Group: Visit:	Screening ID:			

12. What do you think about the amount of physical education lessonsat school?

- _____1 There should be more lessons that at present
- _____2 There should be as many lessons as at present
- _____ 3 There should be less lessons that at present
- 13. How do you find your present fitness?
 - ____1 Very fit
 - ____2 Fit
 - _____ 3 Moderately fit
 - _____4 Not fit at all

14. Do you think that regular exercise is an effective way to keep you healthy?

- _____1 Yes, very effective
- ____2 Quite effective
- _____3 Slightly effective
- _____4 No at all effective