

**Body composition, physical activity and
C-reactive protein in children:
The PLAY Study**

B Harmse, B.Sc. Dietetics

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Abstract

Obesity is currently the most common and costly nutritional problem in developed countries and ten percent of the world's school-aged children are estimated to be overweight to some extent. Low-grade systemic inflammation is increasingly emerging as a significant component of the metabolic syndrome. Youth in lower income families are particularly vulnerable because of poor diet and limited opportunities for physical activity. In developing countries obesity among youth is rising among the urban poor, possibly due to their exposure to Westernised diets coinciding with a history of undernutrition. The aim of this study was to assess the association between serum CRP and physical activity and to assess the association between serum CRP and body composition in black high-school children from a township in the North West Province (NWP), South Africa.

Methods and results: The study group consisted of 193 school children between the ages 13 to 18 years (78 boys and 115 girls) residing in Ikageng, the township outside of Potchefstroom in the North West Province, South Africa. Children were from a black ethnic group, living in a poor socio-economic setting. Demographic and body composition measurements were taken and fasting blood samples were drawn for serum C-reactive protein (CRP) measurements. The difference between serum CRP of overfat versus girls with a normal fat percentage was non-significant ($p = 0.46$). Boys with body fat percentage $>20\%$ ($n=16$) had a mean serum CRP of 1.42 ± 2.16 mg/L and for boys with a normal fat percentage ($n=53$) mean serum CRP was 0.89 ± 1.62 mg/L. The Mann-Whitney U-test for the difference between mean CRP of the two groups of boys was $Z=1.39$, $p = 0.16$ (no significant difference), but with a trend of higher serum CRP concentration in the boys with higher % body fat. For the boys, the only positive partial correlation was between serum CRP and triceps skinfold ($r=0.327$, $p=0.045$). In the girls' group no statistically significant partial correlations were found between CRP and body composition variables. There was no significant difference between serum CRP concentrations of the three physical activity categories of girls. Interestingly, there was an inverse correlation between percentage body fat

and fitness in the boys' group ($r=-0.509$ and $p= 0.008$). The difference in log CRP between activity groups showed a trend of lower serum CRP with higher physical activity in the girls.

Conclusion: This study showed no statistically significant associations between serum CRP and body composition, except for the positive correlation between triceps skin fold and serum CRP in boys, or CRP and physical activity, but clear trends were noted of an inverse association between CRP and physical activity in the girls.

Key words:

C-reactive protein, inflammation, physical activity, youth, adolescent, metabolic syndrome, body composition

Opsomming

Vetsug is tans die duurste en mees algemene voedingsprobleem in ontwikkelde lande, en daar word geskat dat tien persent van die wêreld se kinders van skoolgaande ouderdom tot 'n mate oorgewig is. Laer-graad sistemiese inflammasie word al hoe meer genoem as 'n betekenisvolle komponent van die metaboliese sindroom. Tieners in lae-inkomste families is veral vatbaar omdat hulle minder geleentheid tot fisiese aktiwiteit en minder optimale eetgewoontes ondervind. In ontwikkelende lande is obesiteit in arm stedelike groepe aan die toeneem, moontlik as gevolg van blootstelling aan die Westerse dieet na 'n geskiedenis van ondervoeding in die verlede. Die doel van die studie was om die assosiasie tussen serum C-reaktiewe proteïene (CRP) en fisiese aktiwiteit en ook die verhouding tussen CRP en liggaamsamestelling te bepaal onder 'n groep hoërskool leerlinge uit 'n informele nedersetting in Ikageng, buite Potchefstroom in die Noordwes provinsie van Suid Afrika.

Metodes en resultate: Die studie-groep het bestaan uit 193 skoolkinders tussen die ouderdom van 13 en 18 jaar (78 seuns en 115 dogters) wat woonagtig is in Ikageng, buite Potchefstroom in die Noordwes Provinsie,

Suid-Afrika. Die swart kinders was van 'n lae-inkomste groep. Demografiese en liggaamsamestelling metings is geneem en vastende bloedmonsters is geneem vir serum CRP metings. Die verskil tussen serum CRP van oor-vet teenoor meisies met 'n normale vet persentasie was nie statisties betekenisvol nie ($p = 0.46$). Seuns met 'n vet persentasie $>20\%$ ($n=16$) het 'n gemiddelde CRP van 1.42 ± 2.16 mg/L, en vir seuns met 'n normale vet persentasie ($n=53$) was die gemiddelde serum CRP 0.89 ± 2.16 mg/L. Die Mann-Whitney U-toets vir die verskil tussen die log CRP van die twee seuns groepe was $Z=1.39$, $P=0.16$ (nie betekenisvol), maar daar was 'n tendens van hoër CRP waardes in seuns met 'n hoër persentasie liggaamsvet. Vir die seuns was die enigste betekenisvolle positiewe korrelasie tussen CRP en die triseps velvou ($r=0.327$, $p=0.045$). By die meisies was daar geen betekenisvolle partiële korrelasies tussen CRP en veranderlikes van liggaamsamestelling nie. Daar was geen betekenisvolle verskil tussen CRP vir verskeie aktiwiteitsvlakke in die meisies nie, maar daar was 'n omgekeerde korrelasie tussen vetpersentasie en fisiese fiksheid in die seuns ($r=-0.509$, $p=0.008$). Daar was wel 'n tendens van laer CRP met verhoogde aktiwiteit by die meisies.

Samevatting: Daar was geen statistiese betekenisvolle assosiasies tussen serum CRP en liggaamsamestelling, buiten die positiewe korrelasie tussen CRP en triseps velvou van die seuns, of CRP en fisiese aktiwiteit in hierdie studie nie, maar duidelike tendense word gesien van 'n omgekeerde verwantskap tussen fisiese aktiwiteit en CRP in die meisies.

Kernwoorde:

C-reaktiewe proteïene, inflammasie, fisiese aktiwiteit, jeug, adolessent, metaboliese sindroom, liggaamsamestelling.

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Such confidence as this is ours through Christ before God. Not that we are competent in ourselves to claim anything for ourselves, but our competence comes from God. 2 Corinthians 3:4-5

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Chapter 1: Introduction

Content of introduction

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1.1. Background

South Africa is a developing country where both undernutrition and overnutrition is seen. The prevalence of obesity is high among adult black women, whilst low in children (Kruger, 2005:1153; Steyn *et al.*, 2005). According to Monyeki *et al.* (1999:287), obesity is not only common in South African female adults, but also in female adolescents. In 2002 the First South African National Youth Risk Behaviour Survey stated that the prevalence of overweight among high school children was 17% and obesity 4% (Medical Research Council of South Africa, 2002:12).

Obesity is currently the most common and costly nutritional problem in developed countries with ten percent of the world's school-aged children

estimated to be overweight to some extent (Molnár and Livingstone; 2000:S45). The metabolic syndrome (MS), a cluster of 5 biological markers that together predict the development of cardiovascular disease and type 2 diabetes, is now increasingly emerging among children and adolescents (Nemet *et al.*, 2003:148). Low-grade systemic inflammation is increasingly emerging as a significant component of the MS (Klein-Platat *et al.*, 2005:1178).

Youth in lower income families are particularly vulnerable because of poor diet and limited opportunities for physical activity (PA) (Lobstein *et al.*, 2004: 5). In developing countries, obesity among youth is most prevalent in wealthier sections of the population, but is also rising among the urban poor in these countries, possibly due to their exposure to Westernized diets coinciding with a history of undernutrition (Lobstein *et al.*, 2004: 5). Lambert *et al.* (2004:1762) concluded that the metabolic correlates of excess weight, including a state of low-grade systemic inflammation, are detectable early in life.

This study was performed as part of the PLAY study, which investigated the effects of physical activity in children. The PLAY study (acronym for Physical Activity in the Young) was a parallel intervention study consisting of an experimental as well as a control group with the intervention group having been subjected to a physical activity intervention.

The study group consisted of 193 (78 boys and 115 girls) school children between the ages 13 to 18 years, attending one of two schools (Seiphemelo Secondary School or Boitshoko High School) in Ikageng, a township outside of Potchefstroom in the North West Province, South Africa. A nutrition advisor from the District Health office selected these schools because it was most likely to find undernourished children in these schools.

The sample comprised children from a black ethnic group living in a poor socio-economic setting. The type of housing utilised by the population group is mainly galvanized/zinc or brick houses with a partial water and electricity supply. Subjects in the different schools were in the similar growth phase and socio-economic status and their eating habits and physical activity levels were also similar.

In South Africa there is a need for data regarding the status of childhood overweight, the metabolic syndrome and the cardio-vascular risk, with many studies focusing on only one or two risk factors. In this study, the focus was on gathering information in such a manner that the relevant conclusions could be made regarding body composition, physical activity, CRP as a marker and ultimately the metabolic syndrome and the risk for cardiovascular disease in high school aged children.

1.2. Goals

The goals of the study were

- To assess the association between serum CRP and physical activity in black children aged 14 - 18 from a township in the North West Province (NWP), South Africa
- To assess the association between serum CRP and body composition in black children aged 14 - 18 from a township in the NWP, South Africa.

1.3. Hypothesis

The following hypotheses were formulated for this study:

- There is a negative association between serum C-reactive protein concentration and habitual physical activity in black adolescents from a township school in the NWP, South Africa
- C-reactive protein is positively associated with body composition measures of overweight (especially in terms of body fatness) in black adolescents from a high school in a township in the NWP, South Africa.

1.4. Structure of this mini-dissertation

This mini-dissertation is divided into six chapters. The introductory section is aimed at stating the problem, introducing the reader to the study group and placing the setting of the study in perspective, whilst the literature review is a summary of current peer reviewed literature available on the topic and relevant studies that have been done, followed by the methodology and results obtained. The author concludes with a discussion of results and the significance of the outcomes in light of current literature, followed by the conclusion.

1.5 Contributions of the author

The author played a part in the organisation of the study on ground level and assisted in the collection of demographical data. She also assisted in obtaining anthropometrical measurements (skin folds) and manually computerising data. Interacting with the subjects as well as assisting with interviews on the school premises gave the author the opportunity to attain insight into the circumstances and daily activities of the children.

Chapter 2: Review of literature

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2.1. Introduction

Obesity is currently the most common and costly nutritional problem in developed countries (Molnár & Livingstone; 2000:S45) and ten percent of the world's school-aged children are estimated to be overweight to some extent (Lobstein *et al.*, 2004: 4).

The prevalence of overweight is rising significantly in most parts of the world, although dramatically higher in economically developed regions (Jebb *et al.*, 2003: 461; Lobstein *et al.*, 2004: 4). The rapidity of this increase implicates environmental rather than genetic factors (Lobstein *et al.*, 2004:5), although one cannot exclude the correlation between genes and the environment (Berkey *et al.*, 2003: 836; Guo *et al.*, 2000:1634).

The metabolic syndrome (MS), a cluster of 5 biological markers that together predict the development of cardiovascular disease and type 2 diabetes, is now increasingly emerging among children and adolescents (Nemet *et al.*, 2003:148). Hypertension, insulin resistance, central adiposity, hypertriglyceridemia and decreased values of high-density lipoprotein cholesterol are regularly measured in clinical medicine, but they seem to have little in common mechanistically (Phinney, 2005:115).

Low-grade systemic inflammation is increasingly observed as a significant component of the MS (Klein-Platat *et al.*, 2005:1178). Different cytokines and chemical messengers, which induce their effects individually or in interaction

with each other, constitute the main regulators of the inflammatory process (Klein-Platat *et al.*, 2005:1178).

Among these cytokines, IL – 6, a pro-inflammatory cytokine produced by different cells, adipose tissue amongst others, is over expressed in adults with MS (Visser *et al.*, 2001:e13) and in obese adolescents (Visser *et al.*, 2001:e13). The findings linking inflammation and the MS may either make this picture more complex or provide a mechanistic link between these indexes (Phinney, 2005:115).

Elevated plasma lipid levels are a characteristic of obesity, infection and other inflammatory states. Hyperlipidemia in obesity is in part causal to the induction of peripheral tissue insulin resistance and dyslipidemia contributes to the development of atherosclerosis (Wellen & Hotamisligil, 2005:112)

Youth in lower income families are particularly vulnerable because of poor diet and limited opportunities for PA (physical activity). In developing countries, obesity among youth is most prevalent in wealthier sections of the population, but is also rising among the urban poor in these countries, possibly due to their exposure to Westernized diets coinciding with a history of undernutrition (Lobstein *et al.*, 2004: 5).

2.2. Adolescent physiology and body composition

2.2.1. Physical growth and maturation

Adolescence is an important period in development and significant somatic growth and maturation of secondary sexual characteristics are evident during this time. The onset of puberty is believed to occur as a consequence of a change in the pituitary-gonadal axis resulting in a dramatic rise in testosterone in boys and estrogen in girls. Evaluation of growth based on chronological age alone can be inaccurate and misleading due to marked variability in the timing of maturational changes (Cole *et al.*, 2000:1240; Lobstein *et al.*, 2004:36).

Obese youth characteristically have accelerated growth initially, e.g. in advanced height and bone age, but their pubertal growth spurt is less pronounced, resulting in a reduction in height centile and ultimately adult heights no different from their non-obese counterparts (Lobstein *et al.*, 2004:36).

The pubertal growth spurt is associated with significant changes in body composition (Guo *et al.*, 2000:1633), where girls tend to accumulate more fat than boys. Menarche usually occurs shortly after the peak in height velocity in girls (Kruger, 2005:1153). The rise in serum oestradiol relates temporarily to breast enlargement, widening of the hips and an increase in body fat.

2.2.2. Measures of overweight in childhood and adolescence

Anthropometry is widely used in surveys as an indicator of nutritional and health status. It is especially important during adolescence as it allows evaluation of physical and maturational growth as well as health risks during this critical stage of development (Al-Sendi *et al.*, 2003:367).

Characterisation of BMI-trends and other indicators of body fatness during childhood and adolescence is important so that strategies can be developed to control and prevent overweight and to ensure accurate assessment of body composition (Al-Sendi *et al.*, 2003:368).

BMI as a measure of body fatness in adolescence is influenced by maturation status, race and the distribution of body fat. The relationship between percentage body fat and BMI is dependant on the stage of maturation (for equivalent BMI, lower percentage of body fat in more sexually mature than less sexual mature), race (from an equivalent BMI, whites have a higher percentage body fat than blacks), and waist:hip ratio or waist circumference (for equivalent BMI, central obesity is associated with a higher percentage body fat than peripheral obesity) (Lobstein *et al.*, 2004:36).

Overweight prevalence is significantly higher in early maturers of all racial groups and early maturation is associated with a greater risk of obesity during both adolescence and adulthood (Lobstein *et al.*, 2004:36). Overweight boys

tend to mature later than their non-overweight counterparts. Although early sexual maturation is associated with overweight in girls, in boys the reverse seems to be case, with the prevalence of overweight and obesity higher in late maturers than in early maturers (Lobstein *et al.*, 2004:25).

Cole *et al.* (2000:1243) established the international cut-off points for body mass index for overweight and obesity by sex between 2 and 18 years, obtained by averaging data from Brazil, Great Britain, Hong Kong, Netherlands, Singapore and the United States (Cole *et al.* 2000:1243). For the purposes of this review, data for ages 10 to 18 are presented in Table 1.

2.2.3 Genetic and environmental influences on adolescent growth and development

Population variations in growth are the result of an interaction between genetic and ethnic factors as well as a variety of environmental influences, including socio-economic status and health status (Al-Sendi *et al.*, 2003:374). It is a common impression that school children from non-Caucasian backgrounds living in Westernised societies have a greater propensity for developing obesity than white Caucasian children, but when socio-economic circumstances and parental education are taken into account, the differences may not be great. In the USA for example, African-American and Hispanic Americans appear to contribute more to the obesity epidemic, with more rapid rates of change in their populations than the white American population (Lobstein *et al.*, 2004:42).

Table 1: International cut - off points for body mass for overweight and obesity by sex between 10 and 18 years, defined to pass through body mass index of 25 and 30 kg/m² at age 18 (As defined by Cole et al. 2000:1243)

Age (years)	Body mass index 25 kg/m ²		Body mass index 30 kg/m ²	
	Males	Females	Males	Females
10	19.84	19.86	24.00	24.11
10.5	20.20	20.29	24.57	24.77
11	20.55	20.74	25.10	25.42
11.5	20.89	21.20	25.58	26.05
12	21.22	21.68	26.02	26.67
12.5	21.56	22.14	26.43	27.24
13	21.91	22.58	26.84	27.76
13.5	22.27	22.98	27.25	28.20
14	22.62	23.34	27.63	28.57
14.5	22.96	23.66	27.98	28.87
15	23.29	23.94	28.30	29.11
15.5	23.60	24.17	28.60	29.29
16	23.90	24.37	28.88	29.43
16.5	24.19	24.54	29.14	29.56
17	24.46	24.70	29.41	29.69
17.5	24.73	24.85	29.70	29.84
18	25	25	30	30

2.2.4 Gender differences in adolescent body composition

Sexual development is an important factor influencing anthropometric measurements in body composition during adolescence (Al-Sendi *et al.*, 2003:376). During adolescence, gender differences and age variations become apparent in fat mass, fat-free mass and regional body fat distribution. The rate of increase in BMI is related to high adult BMI levels in both genders during adolescence, but more so in men than in women (Guo *et al.*, 2000:1633). While body fat increases until the age of 17 years in girls, it starts decreasing around the age of 13 in boys.

Adolescence is one of the most vulnerable periods for the development of overweight and obesity (Kruger *et al.*, 2004:564; Lobstein *et al.*, 2004:37). Although the mechanism is unclear, it is possible that fat distribution patterns established during adolescence play a role. Reports suggest that boys have higher WHR values than girls, reflecting a more centralised fat distribution in boys (Al-Sendi *et al.*, 2003:376). Boys tend to deposit fat centrally and lose fat peripherally as they mature, creating a picture predictive of diabetes, heart disease, hypertension and hyperlipidemia in adults (Lobstein *et al.*, 2004:37).

Conversely, lean body mass increases steeply up to the age of 19 years in boys whereas in girls it stagnates at age 15 (Al-Sendi *et al.*, 2003:376). Boys tend to have larger BMI's than girls and the rate of change in BMI is larger in boys than girls (Guo *et al.*, 2000:1634). Interestingly, Guo *et al.* (2000:1634)

found that the maximum BMI attained at post-pubescence is strongly associated with the degree of fatness in adulthood.

2.3. Prevalence of obesity among children and adolescents

The National Health and Nutrition Examination Survey II, conducted in the USA during the period of 1988 – 1994, found that 11% of children and adolescents 6 to 17 years of age were overweight, reflecting a body mass index above the 95th percentile relative to gender and age-specific national reference data (Salbe *et al.*, 2002:299). A report on the initial results of the 1999 National Health and Nutrition Examination Survey indicated that prevalence rates had increased even further in the USA to 13% of children aged 6 to 11 years and 14% of adolescents aged 12 to 19 years (Ford *et al.*, 2001:486; Salbe *et al.*, 2002:299).

South Africa is a developing country where both undernutrition and overnutrition is seen. The prevalence of obesity is high among adult black women, whilst low in children (Kruger, 2005:1153). Stunting is a very common nutritional disorder in South Africa and local research has shown that there may be a link between stunting and the development of overweight or obesity (Jinabhai *et al.*, 2003:358; Kruger *et al.*, 2004:564; Monyeki *et al.*, 1999:287; Mukuddem-Petersen & Kruger, 2004; Steyn *et al.*, 2005:4).

According to Monyeki *et al.* (1999:287), obesity is not only common in South African female adults, but also in female adolescents. In 2002 the First South African National Youth Risk Behaviour Survey stated that the prevalence of overweight was 17% and obesity 4% (Medical Research Council of South Africa, 2002:12). This survey was a cross-sectional prevalence study amongst secondary school learners in South Africa. The study sample comprised of Grade 8-11 (aged 13-19) learners from government schools in 9 provinces in South Africa (Medical Research Council of South Africa, 2002:11), that is 188 schools and 10 699 learners throughout the country. These data also showed that the co-existence of stunting and being overweight is a public health problem among adolescents in SA.

2.4. Consequences of childhood obesity

Many obese children, especially adolescents, tend to stay obese or overweight as adults and it has been suggested that 33% of adult obesity starts in childhood (Forshee *et al.*, 2004:463; Molnár & Livingstone; 2000:S46). Obesity during childhood seems to increase the risk of subsequent morbidity, whether or not obesity persists into adulthood (Graf *et al.*; 2005:291, Molnár & Livingstone; 2000:S46), with obese children being at an increased risk of metabolic and cardiovascular disorders later in life (Jebb *et al.*, 2003; Nemet *et al.*, 2003:148). Generally known obesity-related disorders are heart disease, diabetes, certain cancers, gall bladder disease, osteoarthritis and endocrine disorders and these are on the increase in young adult populations (Lobstein *et al.*, 2004: 4).

One should not forget that the most widely spread consequences of childhood obesity could be psychological (Schwimmer *et al.*, 2003:1813). Schwimmer *et al.* (2003:1818) concluded from a cross-sectional study that severely obese children have lower health-related quality of life (QOL) than children and adolescents who are healthy, and similar QOL to those diagnosed as having cancer.

2.5. C-reactive protein (CRP)

2.5.1 Physiological role of CRP

CRP, synthesized in the hepatocytes, is an acute phase reactant that responds non-specifically (Ford *et al.*, 2001:486) to infections, immunoinflammatory diseases and malignancies (Vikram *et al.*, 2004:1336). It is part of the pentraxin family of ligand-binding and calcium-dependant plasma proteins (Misra, 2004:478).

CRP is also a surrogate marker for IL-6 activity and is proven to predict the development of type 2 diabetes and mortality (Fernandez-Real *et al.*, 2003:1362; Klein-Platat *et al.*, 2005:1178; Vikram *et al.*, 2003:305).

Levels of CRP are usually low or undetected in healthy subjects, but they increase up to 100 times during acute illness or inflammation (Wu *et al.*, 2003:94). In the absence of infection, elevations of CRP levels generally below 10mg/L are associated with an increased risk of the development of

atherosclerotic cardiovascular disease (Cook *et al.*, 2000:140; Sothorn, 2004:704) and in recent years CRP-values, as measured by a high-sensitivity assay (hs-CRP), have been recognised as a useful and sensitive predictor of the future risk of MI and stroke. *De novo* hepatic synthesis starts rapidly after initial stimulus, with serum concentrations rising above 5mg/L by about 6 hours and peaking around 48 hours (Hiura *et al.*, 2003:541).

Pepys and Hirschfield (2003:1805) note that CRP values cannot be used diagnostically, but should be interpreted with full knowledge of all other clinical and pathological results.

An advantage of using CRP is that no fasting is needed before measurement (Genest *et al.*, 2003:5). Duplicate measures, preferably 2 weeks apart are recommended and the lower value should be regarded as the most reliable value (Genest *et al.*, 2003:5, Misra, 2004:478). The plasma half-life of CRP is about 19 hours and is constant under all conditions of health and disease, so that the sole determinant of circulating CRP is the synthesis rate (Pepys & Hirschfield, 2003:1805).

Upon interpretation of CRP-values, low inflammation risk is defined as a level less than 1 mg/L; average risk is 1.0-3.0 mg/L and high risks are values 3-10mg/L (Genest *et al.*, 2003:5; Kushner *et al.*, 2006: 166.e18; Verma *et al.*, 2004:1915). Tests should be repeated and the patient examined for the sources of infection and inflammation (Genest *et al.*, 2003:5).

2.5.2 CRP and Obesity

There is an increasing link between basal inflammation, MS and obesity (Wellen *et al.*, 2005:1112). The release of IL-6 from the visceral (Isasi *et al.*, 2003:332) adipose tissue may induce low-grade systemic inflammation in subjects with increased body fat. This may explain the association between BMI and CRP levels (Wu *et al.*, 2003:97). CRP may be indirectly associated with TNF- α , IL-6 and BMI. These phenomena could also explain why obesity was associated with clinically raised CRP levels in both genders (Wu *et al.*, 2003:98).

Overall adiposity is an important determinant of serum CRP in adults and in children with different ethnicities (Barbeau, 2002:415; Sothorn, 2004:704; Wärnberg, 2004:559). A correlation of CRP with IR, independent of body mass index, has also been reported in children (Sothorn, 2004:704). Vikram *et al.* (2004:1340) found that hs-CRP correlates well with measures of generalised as well as abdominal obesity in adolescents.

It is well known that CRP levels in adults rise with aging, smoking, progression of hypertension and BMI (Hiura *et al.*, 2003:541). It has been proven that weight loss and the improvement of IR leads to decreases of CRP levels and also of event risk in adults. However, the limited number of studies on children causes uncertainty in its clinical significance in inflammation of the young (Hiura *et al.*, 2003:542; Misra, 2004:478; Wärnberg *et al.*, 2004:559).

CRP is also correlated with insulin resistance (IR) in adults (Lee *et al.*, 2004:101; Recasens *et al.*, 2005:112). CRP and obesity is correlated in adults (Maachi *et al.*, 2004:993) and Hiura (2003:541) proved that there are elevated levels of CRP in obese boys.

Results from the Third National Health and Nutrition Examination Survey (NHANES III) showed that CRP concentrations were significantly elevated among children with a BMI > 85th percentile. Excess body weight may be associated with a state of chronic low-grade inflammation in boys and girls (Ford *et al.*, 2001:486). Wu *et al.* (2003:94) also revealed that children in the fourth quartile CRP groups were heavier and had significantly higher BMI's and lower HDL levels than children with non-detectable CRP levels, suggesting that elevated CRP levels might be associated with CVD risk factors in 12-16 year olds. Vikram *et al.* (2003, 305) found that overweight measured by BMI, waist circumference and triceps skinfold thickness correlates with increased CRP. Wörnberg *et al.* (2004:599) confirmed these findings.

The inflammatory properties of IL-6 and tumour necrosis factor (TNF) may play certain roles in the stimulation of acute-phase protein production in the liver, which may regulate plasma CRP levels (Wu *et al.*, 2003:97).

2.6. Dietary fatty acids and inflammation

Although fatty acids (FAs) have been implicated in the development of chronic inflammatory conditions, e.g. insulin resistance and obesity, much research is needed in the relation between insulin resistance, obesity, inflammatory activity and dietary FA's (Fernandez-Real *et al.*, 2003:1362).

Links between adiposity and MS are known, but other factors (like diet) are also thought to contribute (Fernandez-Real *et al.*, 2003:1362; Klein-Platat *et al.*, 2005:1178). Unsaturated FA's and n-3 FAs in particular, are receiving increasing attention as potential anti-inflammatory agents (Fernandez-Real *et al.*, 2003:1362).

Previous ideas on adipose tissue were that it is metabolically inert tissue, serving only as a depot for energy substrate and insulation, but one now knows that it is metabolically functional (Nemet *et al.*, 2003:148). Recent investigations have focused on a family of adipose derived cellular mediators (adipocytokines), including TNF- α and IL-6. The importance of these agents is that they are produced by the fat cells and are known to regulate a host of physiological processes directly tied to carbohydrate and fat metabolism and the development of obesity complications such as diabetes and atherosclerosis (Nemet *et al.*, 2003:148).

Studies of diet or plasma FA composition in children, mostly obese children, focused mainly on lipid variation and not on low-grade inflammation

(Samuelson, 2001:333). Studies on children and young adolescents may furnish new insight into the early mechanisms of MS because they are free of lifestyle confounders such as smoking, drug therapy use and alcohol consumption (Blendea, 2005:1338; Klein-Platat *et al.*, 2005:1178).

Fernandez-Real *et al.* (2003:1366) showed that dietary FA's seem to be highly linked to inflammatory activity. They found this to be especially true in subjects with an increased body fat mass. The percentage of saturated FA's and n-6 FA's were significantly associated with circulating IL-6, whereas the percentage of n-3 FA's correlated negatively with CRP in overweight subjects. A study conducted by Arya *et al.* (2006: 865) among urban Asian Indian adolescents on CRP and dietary fatty acids also showed that saturated FA intake is associated with high CRP values. Sialic acid has been proven to be a marker of obesity-related diseases by acting as an integrated marker of the activity of acute phase proteins (Browning *et al.*, 2004:1004). In the same way as CRP, sialic acid has been shown to be associated with and a predictor of cardiovascular disease and type II diabetes. Interestingly, sialic acid has been proven to predict features of the MS independently of BMI in adult women, but no research has been done in adolescents (Browning *et al.*, 2004:1004).

The associations of the percentage FA's and IL-6 need to be interpreted in the context of the atherosclerotic process, as inflammation in the vessel wall plays an essential part in the initiation and progression of atherosclerosis (Fernandez-Real *et al.*, 2003:1367). Atherosclerosis can be defined as an

immune process initiated by endothelial activation and inflammation which progresses by the involvement of the environmental and genetic factors (Ezgü *et al.*, 2005:1384). Damage to the cell vessel wall leads to endothelial cell disruption, resulting in exposure of the underlying vascular smooth muscle cells.

Endothelial and smooth muscle cells produce IL-6, and IL-6 gene transcripts are expressed in human atherosclerotic lesions. Prospective studies indicate that increased IL-6 and CRP on the one hand and FA composition on the other hand are associated with IR, type 2 diabetes and cardiovascular events. Being overweight modulates the relations of FA's to inflammatory markers (Fernandez-Real *et al.*, 2003:1362).

2.7. Physical activity

2.7.1. Role of physical activity in the development of overweight

Overweight in individuals of any population is the result of long-term positive energy balance (Björntorp, 2001:1006; Molnár & Livingstone, 2000:S46). Energy balance in humans must follow the laws of thermodynamics. The general equation for the energy balance in man is: energy intake = energy expenditure + energy stored (Molnár & Livingstone, 2000:S46). Investigations indicate that the cause of obesity lies in behavioural and environmental changes involving large sections of populations. Studies have shown a negative correlation between PA and body fatness (Molnár & Livingstone,

2000:S45). It was suggested that decreased PA or increasing inactivity is probably the main factor accounting for the reduction in total energy expenditure, leading to positive energy balance and increased prevalence in obesity (Molnár & Livingstone, 2000:S45). Egger and Swinburn (1997:779) concluded that even incidental activity can increase energy expenditure and intensity of activity also plays a role.

Apart from the direct thermogenic effect, exercise increases resting metabolic rate, thermogenic effect of food, fat oxidation and may reduce caloric intake (Molnár & Livingstone, 2000:S53). However, PA should not be seen as the sole agent to induce negative energy balance. It has been suggested that children who engage in regular PA are most likely to become active adults and that there is a behavioural tracking of activity levels from childhood to adulthood (Fulton *et al.*; 2004:581, Molnár & Livingstone, 2000:S53).

The rationale behind the motivation of PA in adolescents is thus to establish exercise as a lifelong habit, teach relevant skills, develop components of physical fitness such as muscle strength, flexibility and endurance, reduce the prevalence of obesity, reduce the risk of osteoporosis and coronary heart disease later in life and increase self-esteem (Fulton *et al.*; 2004:581, Molnár & Livingstone, 2000:S52; Roberts, 2000:35).

2.7.2 Physical activity and metabolic profile

Body fatness and central body fat distribution are related to an adverse risk profile in youth (Ball *et al.*, 2003:392; Al-Sendi *et al.*, 2003:367) and reports suggest that PA exerts a positive effect for risk factors of chronic disease. A higher degree of cardio-respiratory fitness has shown to relate to a healthier metabolic profile in children (Ball *et al.*, 2003:392).

PA may protect against heart disease by improving lipid profile, maintaining blood pressure and controlling body weight (Fulton *et al.*, 2004:581; Molnár & Livingstone, 2000:S52). It is also generally accepted that physically active children have better cardiovascular risk profiles than the non-active ones and that PA plays a key role in the prevention and treatment of obesity through its metabolic effects (Fulton *et al.*; 2004:581; Molnár & Livingstone, 2000:S53). It has long been known that weight and IR is strongly correlated (Reinehr & Andler, 2004:111). PA also improves IR in both obese and non-obese youth and weight loss improves insulin sensitivity and decreases hyperinsulinemia, although obese children who maintain weight loss continue to show elevated insulin levels in spite of improved glucose tolerance (Lobstein *et al.*, 2004:26). Isasi *et al.* (2003:332) showed in a study that mean fitness level was higher in boys than in girls but that CRP levels did not differ between boys and girls and fitness level was inversely correlated to CRP in boys.

2.7.3 Physical activity and urbanisation

Much has been written in the literature on the nutrition transition brought on by the urbanisation and Westernisation of communities over the years (Kruger *et al.*, 2004:565), but a subject receiving very little attention is the "PA transition" (Forshee *et al.*, 2004:463). Physical inactivity is currently reducing the quality of life of tomorrow's adults (Roberts, 2000:33).

Adult weight loss is associated with a reduction in the markers of vascular inflammation and IR (Esposito *et al.*, 2003:1799). Weight reduction in adolescence could slow the progression of metabolic risk factors identified with CVD and type 2 diabetes (Hagarty *et al.*, 2004:481). Berkey *et al.* (2003:839) showed that an increase in total recreational activity over a one-year period was associated with a relative BMI decline in adolescents.

2.8. Clinical recommendations for PA and physical fitness

Public health recommendations often address whole communities at large (macro-environment), whilst clinical community-directed recommendations usually pertain to the individual patient and his/her family (micro-environment) (Egger & Swinburn 1997: 479; Fulton *et al.*, 2004:582).

Overweight youths in particular may need to be targeted by recommendations, as they have a greater risk of developing adverse

cardiovascular disease risk factors and type 2 diabetes (Fulton *et al.*, 2004:582; Reinehr *et al.*, 2004:308). Another concern is that these children are at risk of becoming overweight adults when attaining and maintaining weight loss is difficult and they are at greater risk for adult morbidity and mortality (Ebbeling *et al.*, 2002:473; Fulton *et al.*, 2004:582).

Promoting PA is an increasingly difficult task as PA in the school setting is rapidly fading (Forshee *et al.*, 2004:463), and PA has to be a lifetime pursuit, with foundations being laid early on for behaviours in the future (Daley, 2002:23).

Clinical recommendations have to be seen in relation to assessment and counselling by the physician (Fulton *et al.*, 2004:581). Sedentary lifestyles and poor nutrition challenge children who are predisposed to metabolic disorders (Sothorn, 2004:704). Children with two obese parents have an 80% chance of becoming overweight during their lifetime and, if one parent is obese, their risk declines to 40% (Sothorn, 2004:704). Remarkably, only 7% of children of lean parents are likely to develop childhood obesity (Sothorn, 2004:704).

For moderately overweight children, measures to prevent further weight gain combined with normal growth in height, can be expected to lead to a decrease in BMI. For the seriously obese child, treatment regimes are largely palliative and designed to manage and control rather than resolve the problem. Weight control and improved self-esteem may be achieved, but the child is likely to

remain seriously overweight and at risk of chronic disease throughout his or her life. The clinical management of obese children may require an extended amount of time and the assembly of a professional team including a dietician, exercise physiologist and psychologist in addition to the physician (Lobstein *et al.*, 2004: 6).

General endurance training and sustained activities are among several exercise strategies recommended to prevent or treat obesity. Current recommendations are that youths obtain 20-30 minutes of vigorous exercise each day (Roberts, 2000:33).

Adequate strength is an important part of health related fitness and optimal physiological function for both adults and children. It is recognised for its contribution to improved motor performance, self-image and athletic performance. In addition to improvement in muscle strength, resistance training also increases flexibility, improves physical performance, body composition and cardio-respiratory fitness, reduces serum-lipid and reduces blood pressure (Roberts, 2000:34).

Guidelines for resistance training include:

- Children should be encouraged to participate in a variety of activities that involve repetitive movements against an opposing force.
- Before lifting weights, proper technique needs to be demonstrated.
- Manual resistance training (using a partner) is ideal.
- Slow steady movements should be used (Roberts, 2000:34).

Strong *et al.* (2005:737) performed a systematic review of evidence-based physical activity recommendations for school-age youth and concluded that adolescents should participate daily in 60 minutes or more of moderate to vigorous PA that is developmentally appropriate, enjoyable and involves a number activities.

Losing weight over the short term, but then experiencing a rebound gain in weight, remains the usual experience for the majority of obese children and adolescents (Lobstein *et al.*, 2004: 7). As already mentioned, the psychological aspect should not be ignored as behavioural changes take time to be set in place.

Dietary interventions in combination with exercise programmes have been proven to have better outcomes than dietary modulations alone. Exercise modulation alone without dietary modification is unlikely to be effective, as increased energy expenditure is likely to be matched by increased energy intake (Lobstein *et al.*, 2004: 6). Dietary education is very important in youth for food habits should to a great extent be established at this time (Samuelson, 2001:333).

2.9. Public health requires multi-sectorial action

Rapid urbanisation and Westernisation are the cornerstones of modern day South Africa, and may be causal to the fact that overweight in childhood is or may become a public health issue (Underhay *et al.*, 2003:78). Lobstein *et al.* (2004:44) and Simon *et al.* (2004:S102) have emphasized that children from socio-economically deprived environments in most Western societies have a greater risk of obesity than from more affluent groups.

It is of the greatest importance that the decision makers be pro-active in the setting up of protocols for PA before this public health concern gets out of hand (Al-Sendi *et al.*, 2003:367; Forshee *et al.*, 2004:463). For a majority of obese patients, the first point of contact is the primary care physician or public health nurse (Lobstein *et al.*, 2004: 6).

Policies and actions will have to address all levels of the obesogenic environment (Lobstein *et al.*, 2004: 8). Distances from shops where fruit, vegetables and low-energy density foods are affordable and readily available may all contribute to obesity prevalence (Lobstein *et al.*, 2004:35). Family size, position of the obese child in the family, single parent families and both parent families have all been found relevant to prevalence of childhood obesity in some studies (Lobstein *et al.*, 2004:45).

Problematic social trends influencing obesity have emerged over decades and include an increase in the use of motorised transport and subsequent traffic

hazards to walkers and cyclists, a fall in opportunities for recreational physical activity, an increase in sedentary recreation, multiple TV channels around the clock and greater quantities and variety of nutrient dense foods available. Furthermore, there are rising levels of promotion and marketing of energy dense foods, more frequent and widespread food purchasing opportunities, more use of restaurants and fast food stores, larger portions of food offering better “value for money” and an increased frequency of eating occasions, as well as rising use of soft drinks to replace water, e.g. in schools (Kruger *et al.*, 2005:491; Simon *et al.*, 2004:S102; Underhay *et al.*, 2003:78). Changes in these social trends may require increased awareness by countries of the health consequences of the pattern of consumption as the first step in a strategy to promote healthier diets and more active lives (Lobstein *et al.*, 2004:45).

Children are most vulnerable to social and environmental pressures. They can be encouraged to increase their self-control in the face of temptation and given skills and knowledge to help understand the context of their choices, but they cannot be expected to bear the full burden of their choices. The key element for the prevention of childhood obesity is family involvement. According to Savva *et al.* (2004:456), evidence exists that parental PA and dietary intake patterns are predictive of their children’s risk of obesity.

Neighbourhood policies for safe and secure streets and recreation facilities are frequently lacking, though it is of particular importance in areas with high

crime rates, where it is often unsafe for youths to partake in outdoor activities (Kruger *et al.*, 2004:494).

On a greater scale, many researchers have made suggestions on initiatives that can be followed in order to prevent or treat obesity on a public health level, e.g. intervention should be focused on education and address environmental and social issues which can influence behavioural changes (Kruger *et al.*, 2005:495). Authorities at municipal and regional level should support policies and national and international bodies set standards and provide services encouraging better public health. Commercial practices should promote healthy choices and policies at all levels. All disciplines should be reviewed for assessing their health impact, e.g. public sector supply contracts should comply with health and nutrition policies (Lobstein *et al.*, 2004:8; Simon *et al.*, 2004:S102). Furthermore, all parts of the community should be reached and programmes should be adequately resourced as well as evidence-based and programmes should be sufficiently monitored, evaluated and documented to ensure dissemination and ensure the transfer of experience in order to create continuity (Kruger, 2005:495).

Other issues that have been raised as suggestions for intervention are to obtain public funding of quality physical education and sport facilities to place taxes on unhealthy foods and subsidies for the promotion of healthy nutritious foods, the elimination or displacement soft drinks and confectionaries from vending machines in schools, offering healthier choices; putting restrictions or bans on the advertising and marketing of foods to children and the

assessment of food industry initiatives to improve formulations and marketing strategies (Lobstein *et al.*, 2004: 8).

2.10. Conclusion

Lambert *et al.* (2004:1762) concluded that the metabolic correlates of excess weight, including a state of low-grade systemic inflammation, are detectable early in life. Evidence is emerging that the metabolic syndrome is no longer an “adult” syndrome, but is also a disease of lifestyle in children (Nemet *et al.*, 2003:148).

The current approach to treatment is largely aimed at bringing the problem under control rather than affecting a cure (Lobstein *et al.*, 2004: 7). There seems to be evidence that interventions have limited success. Interventions at the family or school level will need to be matched by changes in the social and cultural context so that the benefits can be sustained and enhanced (Lobstein *et al.*, 2004: 7; Simon *et al.*, 2004:S102). One should, however, remember that a holistic approach should be followed in the prevention and treatment of disease, especially a complex situation concerning public health issues. The truth remains that for optimal mental and physical well-being, nutrient-dense foods and PA are essential (Kruger *et al.*, 2005:1155).

Chapter 3: Methods of study

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3.1. Study design

This study was performed as part of the PLAY study, which investigated the effects of physical activity in children. The PLAY study (acronym for Physical Activity in the Young) was a parallel intervention study, consisting of an experimental as well as a control group, the intervention group having been subjected to a physical activity intervention. For the purposes of this study, the base-line data were used.

3.2. Subjects

Grade 9 (age 13-18 year old) learners from a secondary school in Potchefstroom in the North West Province of South Africa, situated in a low-income area, were included in this study. In total 193 children, boys and girls, were available for inclusion. Sixty subjects attending another school in the same area were also included. Subjects in both schools had similar socio-economic status, they were in the same growth-phase and their dietary and physical activity profiles were similar. The base-line survey was done within the period of 2 weeks.

3.3. Ethical considerations

The PLAY study was approved by the Ethics Committee of the North West University (Potchefstroom Campus) (nr. 04M01). All the Grade 9 students of the Seiphemelo Secondary School and the Boitshoko High School were provided with a permission form that had to be signed by their parents before their inclusion in the study. Permission was also obtained from the principals of the schools. Additional approval was obtained from parents before taking blood samples.

3.4. Measurements

3.4.1. Demographical information

Data regarding age, gender, home language, socio-economic status, housing, educational level and occupation of parents/caregivers, accessibility to water and electricity, smoking status, medical history and general health were obtained by individual interviews, performed in each subject's language of preference.

3.4.2. Body composition and anthropometry

The subjects were measured and weighed in their underwear by trained postgraduate Biokinetics students according to standard methods as described by ISAK (International Society for the Advancement of Kinanthropometry (ISAK, 2001).

Height

The height (cm) of the subjects was taken with a vertical stadiometer to the nearest 0.1 cm. The head of each subject was placed in the Frankfort level (when the orbitals lie in the same horizontal angle as the tragion) and the subject stood upright and stretched out with the buttocks and upperback area against the stadiometer. The height was taken at the highest point of the skull. Measurements were taken twice and the aggregate was noted.

Weight

Body weight was measured to the nearest 0.1 kg on a pre-calibrated electronic measuring scale (Precision Health Scale, A & D Company, Saitama, Japan). The scale was calibrated with a 10kg standardised weight. The subject stood with feet slightly apart and without moving whilst looking forward. Measurements were taken twice and the aggregate of the two measurements was noted. Subjects' body composition was also measured by air displacement plethysmography (BOD-POD, Life Measurement Inc, Concord, CA), which was calibrated at the start of each day's measurements with a cylinder containing standardised volume.

Anthropometrical nutritional status

Anthropometrical nutritional status was defined by z-scores and BMI-for-age. For the calculation of BMI (body mass index) the length and weight measurement was used in the following formula

$$\text{BMI (kg/m}^2\text{)} = \frac{\text{body weight (kg)}}{\text{Height}^2 \text{ (m}^2\text{)}}$$

Circumferences

A flexible steel measuring tape (Lufkin, Cooper Tools, Apex, NC, USA) was used for the measuring of circumference measurements to the nearest 0.1cm. The waist and hip circumferences were measured. The waist-to-hip ratio (WHR) was calculated by dividing the abdominal circumference (measured

around the most narrow part of the abdomen) through the hip circumference (taken at the widest part of the hip).

WHR = abdominal circumference (cm)

Hip circumference (cm)

Skin folds

Skin folds taken included triceps, sub scapula, medial calf, abdominal and supraspinal skin folds. These were measured using a John Bull[®] (British Indicators, London, UK) skinfold caliper to the nearest 0.1mm. The right hand side of the subjects were measured in all cases. Landmarks were drawn first, where after post-graduate Biokinetics students, having obtained a level 2-anthropometric qualification previously took measurements under the supervision of a level 3 anthropometrist. Two measurements were taken for each skin fold and the average of the two was used in further calculations. Most subjects were also measured in the BOD-POD for the measurement of their fat percentage and body composition.

Fat percentage

Fat percentage was measured by air displacement plethysmography (BOD-POD, Life Measurement Inc, Concord, CA). The BOD-POD body composition system uses the principle of whole body densitometry to obtain the amount of fat and lean body mass in the body. After calibrating the BOD-POD, the subjects were shown how to use the thoracic gas volume tubes and the measurement was taken to compensate for lung volume. Body weight (kg)

was taken using a calibrated electronic scale. Body volume (litre) was measured by the BOD-POD. This technique uses the ratio between pressure and volume as explained by Boyle's law. The ratio is used to calculate the unknown volume by measuring the pressure directly. The pressure in both rooms reacts immediately and the size of the difference in pressure represents the relative volume of air in each room. Body density is calculated by dividing the body mass by the body volume. The equation for calculating body density in subjects is as follows:

$$D_b = M / (V_{b_{raw}} + 0.40 VTG - SAA)$$

D_b is equal to the body density and SAA and 0.40 VTG is used to maintain isothermal states. M is the weight of the person and $V_{b_{raw}}$ the measured body volume. From this, the fat percentage, fat mass, lean body mass and lung volume can be obtained. Two measurements were taken and the aggregate used.

The subject was prepared for accuracy of measurement according to the following directions:

- A minimum of tight fitting clothing
- Wearing a swimming cap during measurement
- Removing all jewellery items
- The subject emptying their bladder before testing
- The subject should be relaxed, dry and have a normal body temperature before testing.

The following precautionary steps were taken to take measurements:

- Ensure that the correct height and age is entered
- The subject is asked to enter the BOD-POD and relax
- Minimal movement of the subject within the BOP-POD had to be ensured.

3.4.3. Blood analysis

Qualified nursing practitioners obtained fasting blood samples from the subjects. The *vena cephalica* was used to draw 20 ml of venous blood for the preparation of EDTA plasma and serum. For the preparation of the serum, tubes were left to stand for 30 minutes, whereafter it was centrifuged for 15 minutes by 2000g and 4°C for serum and plasma preparations. The serum and plasma were divided into Eppendorff tubes and frozen at -84°C until analyses were done.

Highly sensitive C-reactive protein (hs-CRP) was measured by immunonephelometry (Cardiophase hsCRP, Dade Behring, 2004) at an accredited laboratory (Ampath laboratories, Pretoria). Control serum was used as an external standard. The mean concentration of the controls was 46.7 mg/L, with a range of 43.2 – 59.0 mg/L and a coefficient of variation of 8.6%.

3.4.4. Usual physical activity

Trained field workers used the Previous Day Physical Activity Recall (PDPAR) to obtain information regarding subjects' physical activity of the previous day (this is a 24 hour recall questionnaire) for the previous weekday and one for one previous day of the weekend. According to this, subjects were rated as being low (1), moderately (2) or highly (3) physically active. The subjects filled the previous day's activities for every 30 minutes of time in an activity chart. The type of activity as well as the intensity of the activity was categorised as high, moderate and low according to its intensity factor. Sketches of low (<3 METS), moderate (>3 METS) and high (>6 METS) were used to explain the classification to the subjects. The MET values of physical activity were taken directly out of the "Compendium of physical activities" (Ainsworth *et al.*, 1993) and the energy expenditure chart of the "PDPAR" was used. A relative energy expenditure value in METs (1 MET = 1 kCal/kg/kg/hour) was allocated for every 30 minute square. The values were used to estimate the total daily energy expenditure from the energy expenditure during specific time periods and in specific activities. The number of 30 minute periods with a MET value equalling 3 METS or more, as well as the 30 minute periods with a MET value equalling 6 METS or more was totalled. Subjects were classified as highly active if one or more 30 minute periods were coded with 6 METS, moderately active when two or more 30 minute time frames were coded with 3 METS and low activity was assigned to those subjects who did not meet the standard of high or medium activity standards (Weston *et al.*, 1997).

3.4.5. Physical development

Tanner staging was used to estimate physical maturity by using a questionnaire. The Tanner scale was used by trained professionals in private rooms to estimate the physical maturity of the boys and girls. Tanner 1 is the questionnaire representing the development of pubic hair in both genders. Classification of Tanner 1 ranges from PH1 (no pubic hair) to PH5 (mature phase). Tanner 2 is the questionnaire that estimates the developing of breasts in girls and genitals in boys. Classification ranges from MA1 (undeveloped breasts or genitals) to MA5 (maturity). Five stages of pubertal growth of body hair and breast or genital development were evaluated to estimate pubertal maturity. Sketches describing the five stages were shown to the subjects and each subject marked his/her own developmental phase (Lee *et al.*, 2006: 346).

3.4.6. Physical fitness test

Cardio-vascular fitness

Cardiovascular fitness was calculated with indirect maximal oxygen uptake (VO_2 maximum) by using a "Bleep-test". The test consists of the subject standing behind a line and jogging to a next line, 20 metres further. The purpose of the test is to estimate the cardiovascular perseverance of the subject. A metronome controlled the amount of time allowed for the subjects to reach the next line. The speed was increased with an increase at each level. Whenever the subject did not reach the level within the allowed time, the test was ended. The previous level reached was noted, for example level

6-1. The "Bleep-test" used for this study was the Australian version (Mullineaux, 2001:41). The results of the Bleep-test were converted to an indirect VO_2 -maximum.

3.5. Procedure

The subjects started at the blood station for taking fasting blood samples. Directly after the blood sampling, the body composition (height, weight, circumferences, skin folds and BOD-POD) were measured, followed by the questionnaires (demographic, Tanner and PDPAR) and at the end of the day, the cardiovascular fitness test ("Bleep-test") was undertaken.

3.6. Statistical analysis

The Statistica computer data analysis software system from Statsoft, Inc. (2004), STATISTICA (data analysis software system), version 7 was used to process accumulated data. Data not normally distributed were transformed logarithmically. Descriptive statistics, Spearman and Pearson correlations were used to analyse data. Descriptive statistics were used to describe characteristics of the subjects. The Mann-Whitney U-test, as well as Kruskal-Wallis tests were performed to assess differences between serum CRP levels of children in categories of habitual physical activity, as well as to compare serum CRP levels of children with normal or low percentage body fat with children with high body fat percentages.

Chapter 4: Results

The sample comprised children from a black ethnic group, living in a poor socio-economic setting. The type of housing utilised by the population group was a mainly galvanized/zinc or brick houses with a partial water and electricity supply. Subjects in the different schools were in the similar growth phase and socio-economic status, and their eating habits and physical activity levels were also similar. (Self-reported physical activity comprised of playing with their friends, walking, playing soccer and watching TV).

Only 5.7% of the children admitted to smoking. Of the children who smoked only one was a girl. The median age of starting to smoke was 15 years (13-17 interquartile range) whereas the median daily tobacco consumption was 6 cigarettes per day (2-10 interquartile range).

Table 2: Descriptive statistics for overfat versus girls with a normal fat percentage

Differentiated on account of fat percentage, girls with fat percentages above 25% were classified overfat, according to Lohman (1992)

	Fat % \leq 25% (n=35)		Fat % $>$ 25% (n=101)	
	Mean	Standard dev	Mean	Standard dev
Age (years)	15.43	1.24	15.55	1.41
Weight (kg)	43.98	5.88	51.80	8.52
Height (cm)	155.63	0.04	155.02	6.67
BMI (kg/m ²)	18.12	2.11	21.53	3.09
Waist circumference (cm)	60.55	4.24	66.28	5.78
Hip circumference(cm)	80.71	5.41	87.83	7.33
WHR	0.75	0.04	0.76	0.05
Triceps skinfold (mm)	10.99	3.87	18.11	5.93
Subscapular skinfold (mm)	9.00	2.95	13.99	5.51
Supraspinal skinfold (mm)	7.52	3.23	12.22	4.96
Abdominal skinfold (mm)	12.82	4.15	22.09	7.11
Biceps skinfold(mm)	6.57	1.74	9.96	4.31
Calf skinfold (mm)	14.08	3.79	20.69	6.62
Fat %	20.60	4.64	32.06	4.74
Muscle mass (kg)	33.57	5.16	34.66	5.28
Tanner stage 1*	3.38 (n=32)	0.87	3.52(n=98)	0.74
Tanner stage 2*	3.28 (n=32)	0.85	3.64(n=98)	0.84
	Median	Interquartile range	Median	Interquartile range
Serum CRP (mg/L)	0.30 (n=35)	0.15, 0.91	0.24 (n=99)	0.20, 1.23

Tanner stage 1* based on pubic hair growth; Tanner stage 2* based on breast/genital development stage (Lee *et al.*, 2006:346)

BMI: body mass index; CRP: serum C-reactive protein concentration; WC: waist circumference; TSF: triceps skin fold

The serum CRP concentrations of the children ranged between <0.2-39.8 mg/L. Children with serum CRP concentrations above 10 mg/L were excluded from further analysis because healthy subjects are expected to have serum CRP values lower than 3 mg/L (Genest *et al.*, 2003:5; Kushner *et al.*, 2006: 166.e18; Verma *et al.*, 2004:1915). High risk serum CRP values are between 3 and 10 mg/L which could indicate signs of acute illness or infection (Cook *et al.*, 2000:140;Sothorn, 2004:704).

Table 3: Descriptive statistics for overfat versus boys with normal fat percentage

Differentiated on account of fat percentage, boys with fat percentages above 20% were classified over fat, according to Lohman (1992)

	Fat % \leq 20% (n=70)		Fat % > 20% (n=15)	
	Mean	Standard dev	Mean	Standard dev
Age (years)	15.88	1.56	15.65	1.44
Weight (kg)	47.96	8.55	53.80	14.46
Height (cm)	161.31	8.66	159.51	10.30
BMI (kg/m ²)	18.32	2.28	20.87	3.69
Waist circumference (cm)	65.11 (n=68)	4.94	69.60 (n=14)	7.12
Hip circumference (cm)	77.70 (n=67)	5.34	84.21(n=14)	8.38
WHR	0.84(n=67)	0.06	0.83(n=14)	0.04
Triceps skinfold (mm)	7.74(n=68)	2.60	14.14(n=14)	6.28
Subscapular (mm)	7.18(n=68)	1.82	12.20(n=14)	8.18
Supraspinal (mm)	5.69(n=68)	2.30	11.60(n=14)	8.22
Abdominal (mm)	9.50(n=68)	4.33	21.32(n=14)	11.01

Descriptive statistics for overfat versus boys of normal fat percentage (continued)

	Fat % ≤ 20% (n=70)		Fat % > 20% (n=15)	
	Mean	Standard dev	Mean	Standard dev
Biceps (mm)	4.74(n=68)	1.16	8.36(n=14)	3.40
Calf (mm)	10.02(n=68)	3.50	16.50(n=14)	4.74
Fat%	16.16	4.21	29.62	4.23
Muscle mass (kg)	40.45	7.49	37.99	10.50
Tanner1*	3.75 (n=69)	0.76	3.40	0.83
Tanner2*	3.58 (n=69)	0.88	3.13	0.83
	Median	Interquartile range	Median	Interquartile range
CRP (mg/L)	0.39 (n=55)	0.15, 0.90	0.59 (n=15)	0.20, 1.65

Tanner stage 1* based on pubic hair growth; Tanner stage 2* based on breast/genital development stage (Lee *et al.*, 2006:346)

BMI: body mass index; CRP: serum C-reactive protein concentration; WC: waist circumference; TSF: triceps skin fold

A SD with such a high value indicates that there were outliers that may be excluded from calculations, such as high serum CRP due to infection rather than low-grade inflammation. Subjects with CRP > 10 mg/L were excluded, namely 3 girls and 3 boys. The descriptive statistics of girls per category of habitual physical activity is portrayed in Table 4.

Table 4: Descriptive statistics of girls per category of habitual physical activity (mean ±SD)

Variable	n	Inactive	n	Moderately active	n	Most active
BMI (kg/m ²)	59	20.43±2.94	35	19.96±3.60	10	20.22±3.02
Bleep test score	58	3.27 ± 0.90	37	3.64 ± 1.31	9	4.47± 0.93
CRP(mg/L)	58	1.97±6.00	38	1.74±2.62	10	0.27±0.12
WC (cm)	60	64.54±5.90	38	63.82±6.42	10	63.35±4.13
TSF (mm)	60	16.60±5.91	38	14.50±4.47	10	14.12±5.13

BMI: body mass index; CRP: serum C-reactive protein concentration; WC: waist circumference; TSF: triceps skin fold

Descriptive statistics for boys per category of habitual physical activity are depicted in Table 5. For the purpose of this study, 'inactive' was defined as children with a score of 1 according to the previous day's physical activity recall (PDPAR), moderately active as a score of 2 and 'most active' as a score of 3, according to the coding for the PDPAR proposed by Weston *et al.* (1997:139).

Table 5: Descriptive statistics of boys per category of habitual physical activity (mean \pm SD)

Variable	n	Inactive	n	Moderately active	n	Most active
BMI (kg/m ²)	18	19.35 \pm 2.58	34	18.56 \pm 2.43	23	19.51 \pm 2.92
Bleep test score	18	6.43 \pm 2.49	33	6.2 \pm 1.75	23	7.00 \pm 1.91
CRP (mg/L)	17	0.54 \pm 0.74	32	3.49 \pm 2.30	21	0.99 \pm 1.60
WC (cm)	18	66.42 \pm 4.95	33	64.81 \pm 5.90	22	66.41 \pm 5.60
TSF (mm)	18	9.12 \pm 4.09	33	8.41 \pm 4.11	22	9.36 \pm 4.94

BMI: body mass index; CRP: C-reactive protein; WC: waist circumference; TSF: triceps skin fold

Most children had low serum CRP concentrations within the normal range (<3mg/L) (Genest *et al.*, 2003:5; Kushner *et al.*, 2006: 166.e18; Verma *et al.*, 2004:1915), with only a few increased serum CRP concentrations up to a

maximum of 39.8 mg/L. The maximum CRP for all the girls was 39.3 mg/L and in the inactive group. The maximum CRP value for this group, but also for the entire group of subjects was 39.8 mg/L for a boy from the moderately active group. After exclusion of children with serum CRP > 10mg/L, serum CRP concentrations of children according to habitual physical activity and body fat percentage were again compared. Because the data were not normally distributed, median and interquartile ranges were also calculated (Table 6).

Table 6: Distribution of serum CRP concentration (mg/L) for boys and girls according to habitual physical activity and comparison of boys and girls with normal fat and high body fat percentage (median, interquartile range)

Physical activity category	n	Girls	n	Boys
Inactive	55	0.33 (0.15, 0.94)	17	0.19 (0.15, 0.38)
Moderately active	38	0.46 (0.26, 1.75)	29	0.59 (0.28, 1.06)
Most active	10	0.22 (0.21, 0.26)	21	0.31 (0.16, 0.95)
Normal body fat %	35	0.30 (0.15, 0.91)	53	0.32 (0.15, 0.79)
High body fat %	74	0.34 (0.20, 1.01)	16	0.50 (0.22, 1.38)

There was no significant difference between serum CRP concentrations of the three physical activity categories of girls. The Kruskal–Wallis value for the difference in log CRP in the girls was $H= 5.18$, with a non-significant p value ($p=0.07$) and for the boys H equalled 6.66, but with a significant difference between PA categories ($p=0.04$).

The difference in CRP between activity groups showed a trend of lower mean serum CRP with higher physical activity in the girls, as seen in Figure 1.

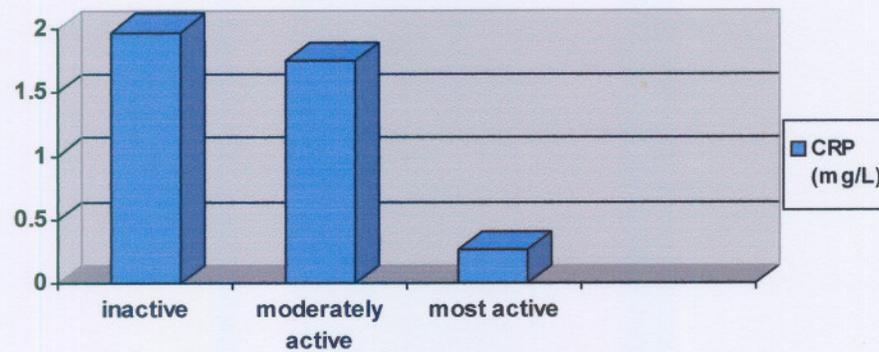


Figure 1: Mean serum CRP levels of girls grouped by habitual physical activity

Comparison of overweight versus lean girls showed the lean group (n = 74) had a median serum CRP of 0.30 mg/L (interquartile range 0.15, 0.9) and the overfat group (n=35) had a median serum CRP of 0.34 mg/L (interquartile range 0.2,1.0)(Table 2). Thus there was no difference in serum CRP between the two groups. Data were also tested for significant difference with the Mann-Whitney U-test for data that are not normally distributed. The difference between serum CRP of the two groups was non-significant, with $p = 0.46$.

Comparing the data for CRP values in boys had the same result, where the boys with increased body fat percentage (n=16) had a median CRP value of 0.5 mg/L (interquartile range 0.2, 1.9) and lean boys (n=53) had a median CRP of 0.32 mg/L (interquartile range 0.15, 0.8) as seen in Table 3. The Mann-Whitney U-test for the difference in CRP in boys was $Z=1.39$, $p = 0.16$,

indicating no significant difference, but a trend of higher serum CRP concentration in the boys with higher % body fat.

Table 7: Pearson partial correlations for boys and girls adjusted for age and smoking (p<0.05 significant)

Parameters	Boys	Girls
BMI and log CRP	0.2085 n=69 p=0.197	0.21 n=105 p=0.116
Waist circumference and log CRP	0.20 n=66 p=0.221	0.19 n=109 p=0.14
Triceps skinfold and log CRP	0.3267 n=66 p=0.045	0.01 n=109 p=0.963
Subscapular skinfold and log CRP	0.2624 n=66 p=0.11	0.07 n=109 p=0.58
Supraspinal skinfold and log CRP	0.233 n=66 p=0.16	0.18 n=109 p=0.16
Abdominal skinfold and log CRP	0.29 n=66 p=0.072	0.12 n=109 p=0.40
Fat percentage and log CRP	0.29 n=50 p=0.151	0.08 n=103 p=0.53
Bleep test at baseline	-0.2828 n=69 p=0.077	-0.18 n=105 p=0.17

Partial correlations attained were performed using the results in Table 2 and Table 3. Pearson correlation analyses were adjusted for age and cigarette

smoking, and subjects with a serum-CRP > 10 mg/L were excluded from these correlations. For the boys, the only significantly positive partial correlation for CRP was between CRP and triceps skinfold ($r=0.327$, $p=0.045$). Interestingly there was an inverse correlation between percentage body fat and fitness in the boys' group ($r=-0.531$ and $p=0.008$) and trends of a correlation between log CRP and abdominal skin folds ($p=0.072$) and bleep test at baseline ($p=0.077$).

In the girls' group no statistically significant partial correlations were found between serum CRP and body composition variables, but trends could be seen of a correlation between log CRP and BMI ($p=0.12$), waist circumference ($p=0.14$) and supraspinal skin folds ($p=0.16$), as well as a trend of a negative correlation between log CRP and bleep test at baseline ($r = -0.178$, $p=0.17$).

Chapter 5: Discussion

The aims of this study were to assess the association between serum CRP and physical activity and to assess the association between serum CRP and body composition in black adolescents in a township school.

It appears that the most active girls had the lowest CRP values, similar to the findings of Isasi *et al.* (2003:332), however, there was no significant difference between the CRP values of active girls versus girls with low habitual physical activity. A definite trend ($p = 0.07$) could however be seen with regards to lower serum CRP in girls with higher habitual physical activity. Physical activity in girls was relatively accurately reported because it is similar to the Bleep-values attained through those methods similar to the methods applied by Mullineaux (2001:41), which measures actual fitness. The reason for the non-significant differences is probably due to the small sample size, with only 10 girls in the most active group.

The results for the boys, however, are difficult to explain, for it seemed that the inactive boys had lower serum CRP values. A possible reason can be that all the boys were relatively fit and active and that other factors were more important determinants of serum CRP concentration. Boys were probably not able to report their PA accurately, also some of the boys might only recently have become inactive. Another reason may be the absence of organised physical activity in rural communities (Underhay *et al.*, 2003:89). It is important to note that this study sample includes only children living in a poor

socio-economic setting, because the literature does state that minor CRP elevation is associated with low socio-economic status and other environmental factors (Kushner, 2006:116e18). The boys' group on the whole, however, (inactive together with active) showed trends as expected (Isasi *et al.*, 2003:332) regarding physical activity and CRP.

As was expected, physical activity was higher in boys than in girls (Isasi *et al.*, 2003:332). Studies have shown a negative correlation between PA and body fatness (Molnár & Livingstone; 2000:S45). In these studies it was noticed, as mentioned before, that more girls than boys were overweight. It has been suggested that decreased PA or increasing inactivity is probably the main factor accounting for the reduction in total energy expenditure, leading to positive energy balance and increased prevalence in obesity (Molnár & Livingstone; 2000:S45). Egger and Swinburn (1997:779) concluded that even incidental activity can increase energy expenditure and intensity of activity also plays a role.

The CRP values for some children were increased above normal concentrations. Upon interpretation of CRP values, low risk is defined as a level less than 1 mg/L; average risk is 1.0-3.0 mg/L and high risks are values 3-10mg/L (Genest *et al.*, 2003:5; Kushner *et al.*, 2006: 166.e18; Verma *et al.*, 2004:1915). In this study, when subjects with markedly increased CRP (>10mg/L) were excluded, a clear trend was observed between activity and CRP in girls (Fig 1). The difference was, however, insignificant probably because of the small sample size in the most active group (n=10). The

relevance of such a study as this in adolescents is useful because CRP is proven to predict the development of type 2 diabetes and mortality (Fernandez-Real *et al.*, 2003:1362; Klein-Platat *et al.*, 2005:1178; Vikram *et al.*, 2003:305). Future risk can thus be assessed and prevented as far as possible.

Smoking is a well-known predictor of serum CRP (Isasi *et al.*, 2003:335), which is why children were questioned on the subject. Only 5.7% of the children admitted to smoking and among the children who smoked, only one was a girl. Adjustment for smoking in correlation analyses did not make any difference to previously non-significant correlations.

A larger number of girls in comparison with boys were classified as overweight. Out of 134 girls, only 33 had a fat percentage equal to or below 25%, with 101 girls having fat percentages above this norm (Lohman *et al.*, 1992). The study group thus probably falls into the typical South African female adolescent profile. According to Monyeki *et al.* (1999:287), obesity is not only common in South African female adults, but also in female adolescents. This also makes sense in light of the pubertal growth spurt, which is associated with significant changes in body composition (Guo *et al.*, 2000:1633) and where girls tend to accumulate more fat than boys. To note differences in body composition is especially important in the context of this study, because adolescence is one of the most vulnerable periods for the development of overweight and obesity (Kruger *et al.*, 2004:564; Lobstein *et*

al., 2004:37). Although the mechanism is unclear, it is possible that fat distribution patterns established during adolescence play a role.

The release of IL-6 from the visceral adipose tissue (Isasi *et al.*, 2003:332) may induce low-grade systemic inflammation in subjects with increased body fat. This may explain the association between BMI and CRP levels (Wu *et al.*, 2003:97). CRP may be indirectly associated with TNF- α , IL-6 and BMI. These phenomena could also explain why obesity was associated with clinically raised CRP levels in both genders (Wu *et al.*, 2003:98). In this study as well, there was no clinically significant difference between values of serum CRP in the boys versus the girls' group.

However, only 15 out of a total of 85 boys had a fat percentage higher than the cut-off percentage of 20%. These results are comparable with current literature (Guo *et al.*, 2000:1633; Molnár & Livingstone, 2000:S45). A positive partial correlation ($r=0.328$, $p=0.044$) was shown between the TSF and serum CRP in boys was shown. This is important because TSF is an indicator of subcutaneous fat, thus boys with more subcutaneous fat are likely to have higher CRP values. This is in line with the literature, which has shown for some time that CRP and overall adiposity are linked (Barbeau *et al.*, 2002:415; Nicklas *et al.*, 2005:1199; Vikram *et al.*, 2003:305; Visser *et al.*, 2001:e13).

Also in line with expectations in the boys' group, there was a trend of an inverse correlation between fitness and fatness (Barbeau *et al.*, 2002:415;

Molnár & Livingstone, 2000:S45). Body fatness and central body fat distribution are related to an adverse risk profile in youth (Ball *et al.*, 2003:392; Al-Sendi *et al.*, 2003:367) and reports suggest that PA exerts a positive effect for risk factors of chronic disease. A higher degree of cardio-respiratory fitness has been shown to relate to a healthier metabolic profile in children (Ball *et al.*, 2003:392).

Rapid urbanisation and Westernisation are the cornerstones of modern day South Africa, and may be causal to the fact that overweight in childhood is or may become a public health issue (Underhay *et al.*, 2003:78). Lobstein *et al.* (2004:44) and Simon *et al.* (2004:S102) have emphasized that children from socio-economically deprived environments in most Western societies, such as subjects of low socio-economic status in this study have a greater risk of obesity than from more affluent groups.

Findings in this study have broad implications, especially with respect to the link between body composition and the risk of cardiovascular risk and the metabolic syndrome. Many obese children, especially adolescents, tend to stay obese or overweight as adults and it has been suggested that 33% of adult obesity starts in childhood (Forshee *et al.*, 2004:463; Molnár & Livingstone; 2000:S46). Obesity during childhood seems to increase the risk of subsequent morbidity, whether or not obesity persists into adulthood (Graf *et al.*, 2005:291; Molnár & Livingstone; 2000:S46) with obese children being at an increased risk of metabolic and cardiovascular disorders later in life (Jebb *et al.*, 2003; Nemet *et al.*, 2003:148).

Conclusion

This study showed no statistically significant associations between serum CRP and body composition or CRP and physical activity, but clear trends were noted of an inverse association between CRP and physical activity in the study sample.

The first hypothesis, that there is a negative association between serum CRP concentration and habitual physical activity in black adolescents from a township school in the NWP, South Africa is thus rejected.

Secondly, the hypothesis that C-reactive protein is positively associated with body composition (especially in terms of body fatness) in black adolescents from a high school in a township in the NWP, South Africa was rejected for girls, as this study showed no statistically significant associations between serum CRP and body composition.

There was, however a positive correlation between TSF and serum CRP in boys, which echoes current literature (Barbeau *et al.*, 2002:415; Molnár & Livingstone, 2000:S45). Also, a trend was noted of an inverse association between CRP and physical activity in the girls confirming that there might be a link between inflammation and physical activity.

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