Graduate School in Experimental and Clinical Nutrition

Nutritional status in evolutive age: assessment of fat mass amount and distribution and of their significance in metabolic derangements

GIACOMO CAGNOLI

2012/2013

SUMMARY

1 INTRODUCTION

1.1 OBESITY

1.1.1 PREFACE

Chronic-degenerative diseases are nowadays in Italy the leading cause of death and requests for health care. Many of them, as indicated by the World Health Organization (WHO), are attributable to behaviors and lifestyles that are established from an early age, including improper eating habits, lack of physical activity and excess weight gain.

Despite the degree of malnutrition on the planet, obesity is a major public health problem in the world. Indeed, we are facing a global epidemic that is spreading in many countries and it can cause, in the absence of an immediate intervention, serious health problems in the coming years.

Obesity is a condition characterized by an excessive accumulation of body fat, usually due to poor diets and a sedentary lifestyle, even if the etiology of obesity reflects a complex interaction between genetic, metabolic, environmental, cultural, socioeconomic, and behavioral factors.

1.1.2 DIAGNOSIS

Fat mass evaluation

An ideal measure of the visceral fat should be accurate, precise, easy, acceptable and well documented. Body fat expressed as percentage of total body weight is difficult to measure in the everyday practical clinic, and shows important variations in correlation to age and sex. The goal is to find a well-documented methodic with sharp cut-offs in order to correlate different populations. The most common used methods in the clinic practical are Body Mass Index (BMI) and waist circumference (WC): they are not direct measures of adiposity, but they have a strong correlation with other specific measures of body fat mass and are predictors of metabolic risk.

Among them, the Body Mass Index is currently the most used; it is calculated by dividing the subject's mass by the square of his or her height (kg/m^2); the interpretation of the BMI suggested by WHO in 1997 is listed in table below.

BMI	Classification
< 18.5	underweight
18.5–24.9	normal weight
25.0–29.9	overweight
30.0–34.9	class I obesity
35.0–39.9	class II obesity
≥ 40.0	class III obesity

In contrast to what happens to the adult BMI values in childhood are constantly changing depending on gender and age. BMI in children is calculated the same way as adults, but compared to typical values for other children of the same age (Cole BMI charts, Cole et Al., 2000): according to the American Academy of Pediatrics, people under 5th percentile for BMI are considered to be underweight, people between 85th and 95th percentile are considered to be overweight, while children over 95th percentile are considered to be obese.
Waist circumference estimates visceral adipose tissue and correlates with the risk of metabolic disease even in children. Today the most widely used index is the waist to height ratio (high values > 0,5), more immediate and simple compared to the value of percentile or z-score of waist circumference.

Anamnestic evaluation
Each patient must be subject to a thorough medical history for a correct diagnosis. The family history detects weight and height of parents and brothers: the presence of obesity in a first-degree relative is one of the most important risk factors for the onset of obesity in children and increases the risk of persistence of obesity with the growth. The

presence of diseases and cardiovascular risk factors in first-degree relatives should be investigated, as well as the presence of diabetes mellitus, endocrine disorders or eating disorders.
The personal history detects problems related to pregnancy (including mother's weight gain during pregnancy), birth weight (very high or very low weight at birth are risk factors for the maintenance of excess weight in later years), the trend of height and weight growth and psychomotor development (its delay may reflect the presence of syndromic obesity), an early adiposity rebound, any regular use of medications and breathing or sleep-related problems.
The behavioral history detects the mode of feeding (breastfeeding or bottle feeding) and the weaning period (as noted, precocious weaning, hyperalimentation and excess of protein intake lead to the development of obesity), a qualitative and quantitative assessment of the risk behaviors of the child and the family, including information regarding nutrition, physical activity and sedentary lifestyle.

Clinical evaluation
The physical examination should be carefully conducted: it must therefore investigate weight, height, BMI and waist circumference, skinfold thickness and height percentile of the annual rate of growth (for evidence of short stature or stunting, generally not compatible with a diagnosis of essential obesity), arterial blood pressure, appearance of genital and pubertal stage, the presence of dysmorphic features or abnormal psychomotor development and any other clinical features such as the presence of acanthosis nigricans, striae rubrae, hypertrichosis (which are correlated with endocrine etiology).

Instrumental and laboratory evaluation
The American Heart Association and the American Academy of Pediatrics recommend an assessment of blood parameters in overweight and obese child after two years of life. These investigations include the assessment of first level like glycemic profile, lipid profile, liver profile; among investigations of second level, assessment of insulin resistance

and liver ultrasound are recommended. In case of detection of fasting hyperglycemia, glycosylated hemoglobin and oral glucose tolerance test (OGTT) are recommended. If there are any signs or symptoms correlated to secondary obesity, complications related to the disease in question will be investigated

1.1.3 DIFFERENTIAL DIAGNOSIS

As mentioned before, it is always easier to find excess weight in young children. It's 'well known that most of the times we are faced with a case of essential obesity (about 95% of cases); however, the early onset of obesity in children may be due to a secondary form, underlying causes that have to be carefully investigated and excluded.

Endocrine obesity

Endocrine dysfunctions that play a pathogenic role in obesity of early onset are rare (less than 1% of all obesity), but they must be recognized early in life in order to allow an adequate and timely treatment. In these cases, the excess weight is generally linked to signs and symptoms that may alert clinicians to the correct diagnosis.

The clinical or subclinical hypothyroidism is relatively common in the first months or years of life. In cases of clinical hypothyroidism, it can easily show symptoms related to the disease (such as dry skin, constipation, somnolence, apathy, hyporeactivity...); the dosage of thyroid hormones is easily able to confirm the diagnosis.

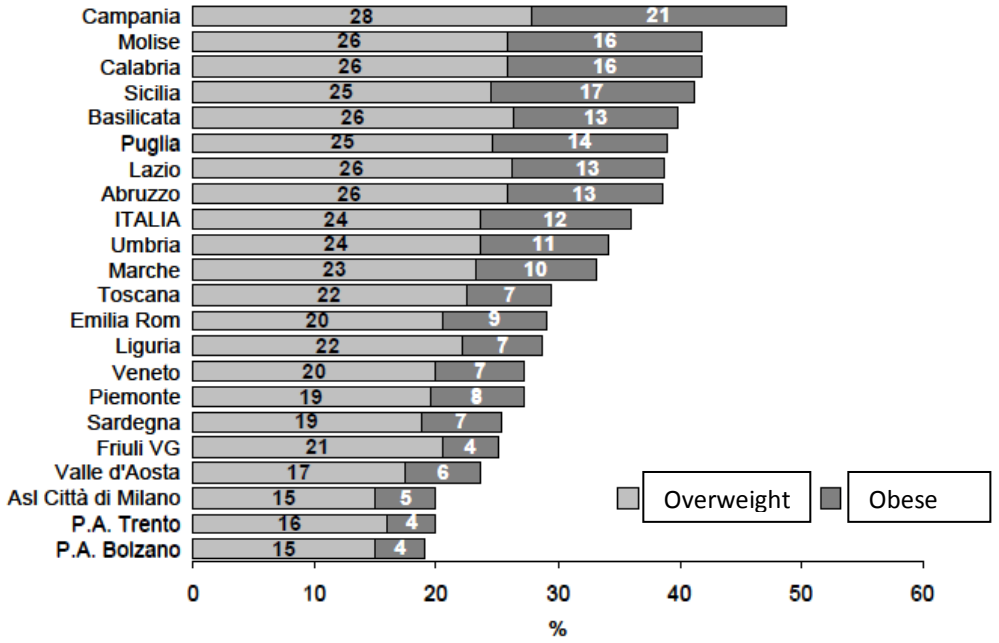
The hyperadrenocorticism, either by adrenal and hypothalamic-pituitary origin (Cushing's syndrome and disease), is very rare in the early stages of life; Cushing's syndrome is most often iatrogenic, due to abuse of preparations of steroids. This disease should always be suspected in the presence of a low stature with growth retardation of bone age, tendency to accumulate fat in the trunk and abdomen, the presence of striae rubrae in the abdominal region, hips, thighs and mammary region and blood pressure higher than normal. Basal adrenal function tests and high urinary free cortisol help to direct a possible adrenal dysfunction.

The presence of early onset obesity in the first years of life can also be the first sign of an organic disease of the hypothalamic-pituitary region (craniopharyngioma and pituitary adenoma), when the excess weight has risen rapidly and is associated with sudden slowdown in growth velocity or signs of intracranial hypertension. In these cases, an imaging examination such as MRI is able to confirm or rule out the suspected diagnosis.
Among the more rare forms of obesity etiology endocrine, we recall the presence of pseuohypoparathyroidism, GH deficiency and nesidioblastosis.

Genetic obesity
The presence of obesity in a child associated with other features such as mental retardation, short stature, cryptorchidism or hypogonadism, dysmorphic features and other changes, can lead to think of a form of obesity on a genetic basis, which must be supported by specific investigations in the diagnosis.
Prader-Willi syndrome is the most frequent of these conditions (with a prevalence of 1:20000) and is due to an alteration of chromosome 15 of paternal origin. In the first months of life the diagnostic suspicion is supported by reduced fetal movements, neonatal hypotonia, lethargy, weak cry, difficulty in feeding and psychomotor retardation; the suspected diagnosis must be confirmed with the methylation test. In these patients obesity is evident after 2-3 years of age and is accompanied by hyperphagia, mild to medium mental retardation, hypogonadism.
Another form of rare genetic disease related to obesity is the Bardet-Biedl syndrome (prevalence 1:100,000), with an autosomal recessive transmission, which is characterized by excess weight associated with visual disturbances (very frequently retinitis pigmentosa), or alterations of color vision, often in the presence of polydactyly. Other forms of syndromic obesity include Cohen Syndrome and Syndrome Alstrom.

1.1.4 EPIDEMIOLOGY

Worldwide there are over 155 million overweight or obese children, and the prevalence of obesity has remarkably increased over the past two decades. The number of overweight children in EU countries is expected to rise by 1.3 million per year, with more than 300,000 children becoming obese each year without urgent action to counteract the trend (Toselli et Al, 2012). In Italy, in 2005 on a sample of 60 000 families, 34% of adults was overweight and 10% obese, showing an increase of 9% compared to the values obtained in 2000; as regards children, in a research conducted in 2008-2009, 23,2% were considered overweight and 12% were obese, with great differences among southern regions if compared with northern regions (Spinelli et Al., 2012).



This trend leads to serious consequences on the health status of the population, with a progressive increase in patients with diabetes, cardiovascular disease and cancer because obesity and overweight are one of the main risk factors for other chronic diseases including type 2 diabetes through the promotion of insulin resistance, cardiovascular diseases, hypertension and stroke and certain forms of cancer (Agostoni et Al, 2011). Consequently it will bring serious consequences on the

financial statements of the States, affecting economic and social development both for the rising costs of health care (with more than 6% of the costs in each European country), and reducing productivity in work and income. It has been estimated that in 2030, the health costs associated with obesity-related diseases will rise of 48-66 billion of dollars only in the United States. An early identification of subjects at risk could can prevent the development of those pathologies, ensuring better quality of life. The problems related to overweight and obesity must be addressed according to a cross-sectional approach, paying attention not only to the aspects specifically linked to health, but also environmental, social and economic determinants. The prevention strategy should start from an early age, because early adiposity accumulation (in the first 5 years of life) is one of the individual risk factors for the development of obesity.

1.1.5 COMPLICATIONS

It's well known that in the adult subjects obesity predisposes to chronic diseases and complications that can lead to increased mortality compared with age-matched normal weight subjects. However, in the child we cannot speak of a real risk of mortality, but given the high persistence of pediatric obesity in adulthood, it is fair to assume that the changes have arisen in the early ages of life are correlated with morbidity in the long term. In Western countries, the adult mortality is mainly due to cardiovascular diseases: the main risk factors are high blood pressure, atherosclerosis, dyslipidemia, and diabetes mellitus. All this metabolic abnormalities are commonly associated with obesity and are known to regress with weight loss. The obese child after a few years presents some metabolic abnormalities: first of all are insulin resistance, impaired glucose tolerance and type 2 diabetes.

Insulin resistance is a condition characterized by the inability of insulin to adequately stimulate the entrance of glucose into the cells; the result is a decreased utilization of glucose that accumulates in the circulation and further stimulates the secretion of insulin. In a first phase, hyperinsulinemia favors the cellular uptake of glucose, which is still

able to maintain normal blood glucose values. Subsequently, blood glucose levels become progressively more pronounced with further increase in insulin secretion: in this phase appears glucose intolerance. Finally, the appearance of high postprandial blood glucose values, which do not result in an adequate pancreatic response, leads to the onset of diabetes mellitus type 2; the lower peripheral glucose utilization also results in a down regulation of insulin receptors and a lower use of liver glycogen.

The metabolic syndrome is a clinical situation frequently encountered in obese adults, characterized by the presence of insulin resistance, glucose intolerance, dyslipidemia and hypertension. This condition is associated with morbidity and mortality from cerebral vascular diseases, cardiac and peripheral devices. The child suffering from obesity can also develop metabolic syndrome, although the young subject shows less clear features.

With regard to dyslipidemia, obese subject presents circulating levels of free fatty acids higher than in normal-weight person, due to the inability of insulin to inhibit their release into the circulation. High blood pressure is not common in obese child, although his pressure values are significantly higher than in normal-weight person; on the other hand, insulin resistance and hypertension are strictly associated in adults.

The circadian rhythm of cortisol is normally preserved, but elevated levels of excretion of metabolites of cortisol and corticosterone can be documented, resulting in increased secretion of ACTH and increased production of adrenal androgens, that can lead to early adrenarche in these children. The child with essential obesity usually does not present growth failure: during the prepubertal period they are often taller than their peers. The adrenarche is usually anticipated, but sexual development is normal. In the female instead menarche is often anticipated, often followed by changes in the menstrual cycle such as secondary amenorrhea and dysmenorrhea. Relatively common is polycystic ovary syndrome, characterized by the presence of numerous ovarian cysts with a diameter greater than 4 cm or oligomenorrhea

associated with dysmenorrhea, signs of hyperandrogenism and hyperinsulinemia.

In children with important obesity drowsiness, hypercapnia, hypoxia and congestive heart failure secondary to pulmonary hypertension are described. The limitations resulting from the decrease in lung compliance commonly lead to easy fatigability and dyspnea during the practice exercise.

Other long term complications strongly linked to obesity are known to be musculoskeletal diseases such as arthrosis and chronical orthopedic problems, nefrosic syndrome and oncological diseases.

1.2 METABOLIC SYNDROME

Metabolic syndrome is defined as a cluster of risk factors for cardiovascular disease and type-2 diabetes mellitus occurring in the same individual; it includes elevated blood pressure, atherogenic dyslipidemia (raised triglycerides and lowered HDL-cholesterol), raised fasting glucose and abdominal obesity (Pacifico et Al., 2013). The syndrome is associated with a two-fold increase for the risk of cardiovascular diseases and a five-fold increase for the risk of type-2 diabetes mellitus. Therefore, it is alarming to see that the prevalence of metabolic syndrome is reaching epidemic proportions worldwide. Moreover, MS typically regarded as a middle to late adulthood disorder, is now emerging in adolescence with close to 10% of all 12-19 years old being affected. There is no universally accepted definition of MS in children and adolescents (Zimmet et Al., 2007; Goodman et Al., 2007). The International Diabetes Federation and American Heart Association have both recently proposed a revised definition of MS in children and adolescents (see table below). In both guidelines, waist circumference was assessed by percentiles, and the cut-offs for metabolic and blood pressure variables were defined only for children above 10 years of age.

Table 1 Definition criteria for metabolic syndrome in children and adolescents.

Ages (years)	AHA criteria[a]	IDF criteria[b]		
	12–19	6–9	10–15	>15 (adult criteria)
Waist circumference	≥90th percentile for age, sex, and race/ethnicity	≥90th percentile for age (MS as entity is not diagnosed)	≥90th percentile or adult cut-off if lower	≥94 cm for Europid males, ≥80 cm for females
Blood pressure	≥90th percentile for age, sex, and height		Systolic ≥130 or diastolic ≥85 mm Hg	Systolic ≥130 or diastolic ≥85 mm Hg or treatment of previously diagnosed hypertension
Triglycerides	≥1.23 mmol/L (≥110 mg/dL)		≥1.7 mmol/L (≥150 mg/dL)	≥1.7 mmol/l (≥150 mg/dl) or specific treatment for high triglycerides
HDL-C	≤10th percentile for race and sex		<1.03 mmol/L (<40 mg/dL)	<1.03 mmol/L (<40 mg/dL) in males and <1.29 mmol/L (<50 mg/dL) in females or specific treatment for low HDL-C
Fasting glucose	≥5.6 mmol/L (100 mg/dL)		5.6 mmol/L (100 mg/dL)	5.6 mmol/L (100 mg/dL) or known T2DM

AHA, American Heart Association; IDF, International Diabetes Federation; HDL-C, high-density lipoprotein cholesterol; T2DM, type 2 diabetes mellitus.

[a] For the diagnosis of metabolic syndrome, 3 of the five must be present.

[b] For the diagnosis of metabolic syndrome, central obesity and 2 of 4 other components must be present.

The dyslipidemia associated with metabolic syndrome consists of both high triglycerides and low high-density lipoprotein cholesterol (HDL) levels, the so called atherogenic dyslipidemia (Bacha F et Al., 2003). Obesity and insulin resistance are thought to promote atherogenic dyslipidemia by enhancing the hepatic synthesis of the triglyceride-rich very low-density lipoproteins (VLDL). In addition, the activity of lipoprotein lipase e the enzyme that catabolizes VLDL and generates HDL particles is impaired in the insulin resistant state, further contributing to raise triglycerides and lower HDL levels (Steinberger J et Al., 2009).

The primary treatment for the dyslipidemia associated with metabolic syndrome is behavioral: weight management and exercise. This approach is strongly supported by evidence indicating that obesity during childhood is related to unfavorable changes in plasma triglycerides and HDL, and that fitness may enhance clearance rate of plasma triglycerides and production of HDL particles.

Glucose abnormalities, defined as impaired fasting glucose or impaired glucose tolerance, represent, by definition, a major component of metabolic syndrome. As with adults, they are strongly related with obesity and insulin resistance in children. Lifestyle modifications including dietary changes and improvements in physical activity are the mainstay of treatment of glucose abnormalities in children with metabolic syndrome. Whole-grain intake has been associated with greater insulin sensitivity and lower BMI in adolescents. Fiber in particular attenuates postprandial glycemic excursions and has beneficial effects on insulin sensitivity, adiposity, and pancreatic function. Children should be encouraged to add at least five fruits and vegetables per day, and minimize or eliminate sweetened beverages. Most importantly, high-fat and high-calorie food items should also be minimized. Physical activity is considered a cornerstone in the prevention of diabetes mellitus type 2 by improving insulin sensitivity and through direct effects on glucose uptake in the skeletal muscle. Recent studies in adolescents have shown that physical activity is

positively associated with improved glucose metabolism and resting energy expenditure and negatively associated with IR-dependent metabolic parameters (Sinha et Al., 2002).

The prevalence of hypertension among obese children is estimated to be as high as 11% (Sorof et Al., 2004). Diastolic blood pressure has been shown to be a function of increasing abdominal circumference. A successful treatment of hypertension in children and adolescents with metabolic syndrome requires a multifaceted approach, beginning with non-pharmacological measures, including weight loss, exercise, and dietary modifications: Studies in obese adolescents have demonstrated that moderate weight loss decreases blood pressure, either systolic and diastolic (Reinehr T, Andler W, 2004). Pharmacologic management is reserved to pediatric patients who do not respond to a lifestyle modification.

1.2 VISCERAL FAT

Epidemiologic evidence supports the theory that the relation between obesity and disease risk begins early in life. For example, in young adults who died in accidents, fatty streaks in the coronary arteries and aorta that were found at autopsy were associated with blood lipid profile, blood pressure, and obesity status obtained at one or more points antemortem (Tracy et Al., 1995). Additional longitudinal data indicate that the occurrence of overweight, hypertension, and dyslipidemia in young adults (aged 19–32 y) was associated with these same risk factors in childhood (Pinhal-Hamiel O et Al., 1996). The evidence indicates that obesity-related disease can begin in childhood and that risk factors for disease track, or remain at a similar level, with advancing age, growth, and development. The relation between obesity and disease was noted as early as 100–200 BC by Samhita and Ayurveda, who observed the relation between glycosuria, obesity, and lifestyle. In 1717, Morgagni noted the android fat pattern in the corpse of a woman and in 1947, Vague described the metabolic risk of android obesity as compared with the protective nature of gynoid obesity (Vague et Al., 1988).

Adipose tissue stores are heterogeneous with respect to metabolic activity and relation to disease risk. Deeper analysis have shown that, not only the total, but the distribution of excessive adipose tissue seems to determine the development of these comorbidities and, therefore, abdominal obesity is pointed out as more harmful to human health than subcutaneous adipose tissue. The specific fat deposit most related to negative health effects is visceral fat (VF), also referred to as intra-abdominal adipose tissue mass IAAT). Visceral fat is metabolically unique when compared with subcutaneous adipose tissue. The increased sensitivity to lipolytic stimuli like norepinephrine and its proximity to the hepatic portal vasculature are hypothesized to be responsible for the positive association between visceral fat, dyslipidemia, hyperinsulinemia, and glucose intolerance. By contributing free fatty acids to the liver, visceral fat leads to increased

circulating triglycerides (TG), decreased high density lipoprotein, increased hepatic glucose production, and decreased hepatic insulin extraction, hence predisposition to metabolic syndrome. Thus reductions in waist circumference, in addition to overall obesity, should be a target for intervention to reverse obesity-related health risks in general population. Essentially, all successful programs include interventions to reduce calorie intake and increase physical activity. Dietary modification should be age-specific, providing appropriate optimum nutrient intake for the maintenance of healthy linear growth and normal development. Recommendations include a moderately reduced calorie intake while maintaining a well-balanced diet. The increase of physical activity must be also considered as part of the treatment, based on the mounting evidence in adults which demonstrates the benefits of regular physical activity on abdominal obesity. However, the role of regular physical activity alone (e.g., without calorie restriction) on abdominal obesity, and in particular visceral fat, is largely unclear in youth. There is some evidence to suggest that engaging in higher-intensity physical activity is associated with a lower waist circumference and less visceral fat. Randomized controlled studies have also shown that aerobic exercise is protective against age-related increases in visceral adiposity in growing children and adolescents. However, evidence regarding the effect of resistance training alone as a strategy for the treatment of abdominal obesity is lacking and warrants further investigation.

The commonly used imaging modalities to measure visceral fat are ultrasonography (US), computerized tomography (CT) and Magnetic Resonance Imaging (MRI). With these approaches, adipose tissue is measured in terms of cross-sectional area (cm^2) or volume (cm^2). Because these techniques are expensive and CT involves radiation exposure, intra abdominal adipose tissue is often measured in a single, cross-sectional slice at an anatomic landmark, usually the level of the umbilicus or the L3–L4 disk space. The major advantages of these imaging techniques are the high resolution of the images and the capability to identify small deposits of visceral fat. In addition,

subcutaneous abdominal adipose tissue (SAAT) is also accurately quantified at the same time. Indirect measures of intra abdominal adipose tissue also include dual energy X-ray absorptiometry (DXA) to measure fat mass in the trunk region and anthropometric measures (circumferences and skinfold thicknesses). Generally speaking computed tomography is considered as the gold standard for visceral fat measurement, but because of the radiation problem and economic reasons, ultrasonography is often preferred for visceral fat estimation in children. In adults the waist-to-hip ratio and waist circumference are often used as markers of visceral fat, even if WC reflects not only the quantity of fat but also the quantity of lean body mass (muscles, bones and internal organs) and cannot distinguish between subcutaneous and visceral fat. As such, waist circumference may misclassify visceral obesity: there are individuals who have a normal WC but an excessive amount of VF and high risk for metabolic syndrome, and individuals who have a large WC but a normal amount of intra abdominal visceral fat and low risk for metabolic syndrome. However, in children and adolescents the correlation between these markers and visceral fat as measured by imaging techniques is not strong. A close relationship between high morbidity and intraabdominal (visceral) fat obesity, rather than extra-abdominal (subcutaneous) fat obesity, has been observed in adults in several studies. Although body fat patterning has been related to adverse health outcomes in adults, its importance in children and adolescents is less certain. If visceral fat accumulation rather than general fat accumulation is a more direct predictor of human cardiovascular disease in adulthood, then an early measure of this fat deposit may be useful in identifying an individual at increased risk. Some studies suggest that IAAT in children increases in proportion to overall fatness as measured in adults, whereas other studies showed that obese children tend to accumulate subcutaneous fat and not IAAT. The influence of dietary factors on visceral fat accumulation has not been extensively studied in children. Physical activity indeed is an important determinant of intra abdominal adipose tissue.

Growth during childhood is known to be a time of rapid change in body composition; however, there have been few longitudinal studies that examined changes in specific fat compartments during the growth process. The study of change of various compartments of fat is important because it helps to elucidate the dynamics of growth in children and how changes in body composition may be related to health outcomes. This is especially important for the growth of visceral fat, which may contribute to metabolic disease risk. In children, visceral fat has been shown to be positively related to a wide range of health indicators, including total cholesterol, low-density lipoprotein cholesterol, triacylglycerol, insulin areas after an oral glucose test, basal insulin secretion, and stimulated insulin secretion; In addition, visceral fat seems to have a negative relationship with insulin sensitivity and high-density lipoprotein cholesterol. Although some associations between different adipose tissues and risk factors have been identified, how visceral fat or subcutaneous abdominal fat is acquired in children and the mechanisms through which visceral fat or subcutaneous abdominal fat affect disease risks are still unclear: there have been few previous reports from longitudinal studies that examined the growth trajectory of visceral fat or subcutaneous abdominal fat in children.

With the use of imaging techniques for direct measurement of intra abdominal adipose tissue, several Authors (Kissebah et Al., 1982; Després et Al., 1984; Björntorp, 1992; Vague et Al., 1988) reported in the 1980s and early 1990s that excess visceral fat explains the relation between obesity and metabolic complications in adults, but first studies conducted in children start to be published in nineties: the first study that has investigated the distribution of body fat in subjects in childhood is from de Ridder et Al in 1992., in which the Authors postulated waist circumference to be a good good measure for the related amounts of fat in pubertal girls. Differences in body fat accumulation have been noted with boys having more visceral abdominal tissues during evolutive age than girsl (Kotani et Al., 1994, Goran et Al., 1995). Goran concluded his study supporting that individual trunk skinfold measurements and the trunk/extremity skinfold ratio provide a better indication of IAAT

compared to the waist/hip ratio, even if the relationship between intra-abdominal adipose tissue and anthropometry in children is complex. First study to investigate the relationship between viscerl fat accumulation in evolutive age and metabolic derangements has been published in 1995 (Caprio S et Al., 1995), where the Authors noted that in obese girls, visceral fat, but neither waist-to-hip circumference ratio nor subcutaneous fat, was highly correlated with basal insulin secretion, stimulated insulin secretion, and insulin resistance. On the other hand, Iwata in his study concluded that visceral fat obesity is a rare status and has no close relationship to coronary risk factors in childhood (Iwata F et Al., 1995), while Ellis in 1997 published a paper in which underlinewd the role of VAT as a potential early marker for cardiovascular diseases (Ellis KJ, 1997). In the following years publication about visceral fat in children and its relationship with other anthropometric measurements and metabolic derangements have been published, but still there's not univocal consent about it.

Nutritional status in children can be evaluated by means of simple anthropometric assessment methods. Body Mass Index, an index of excess weight relative to height, has a moderately high sensitivity for the general identification of the obese child, but a weak correlation with total body fat due to considerable changes in fat mass that occur during childhood. Other simple body composition assessment methods are needed to investigate the metabolic risk for young patients.

2 AIMS OF THE STUDY

In this study we wanted to see the correlation between anthropometric measures and visceral fat assessed by ultrasonography in a large population of children; furthermore we wanted to see the clinical correlation of visceral fat accumulation in childhood obesity with respect to the development of metabolic derangements, compared to body mass index and waist circumference. At last, we wanted to investigate the effects of visceral fat accumulation in developmental age; in particular we supposed that, like in adults, the increment of abdominal visceral fat, measured by ultrasonography, is associated with a worsened metabolic profile and with the increment of metabolic syndrome risk.

3 RESULT AND DISCUSSION

3.1 MATHERIALS AND METHODS

This study was undertaken in The International Center for the Assessment of Nutritional Status (ICANS) of Milan, Italy.
We considered a sample of 83 patients who spontaneously referred to ICANS for valuation of nutritional status and dietary treatment in case of overweight, obesity or nutritional counseling divided in 58 females and 28 males, with age range 11-17 and mean age of 14,6 years. None of them was affected by acute or chronic disease; those with overweight syndromes, overweight secondary to underlying disorders, those with eating disorders (either over or underweight) and those on longterm drugs were excluded from the study. The history was obtained from the parents/guardian and physical examination was performed as per the protocol.
All the patients were subject to anthropometric measurements of weight, height and BMI. Body weight and body height were measured to the nearest 0,1 Kg and 0,5 cm respectively; they were measured while the subjects were fasting and wearing light clothes without shoes. Weight was assessed with a balance-beam scale and height with a stadiometer; for the statistical analysis we calculated z-score for both weight and height.
Body Mass Index was calculated as weight (kilogram) divided by square of height (meter), and then for every patient z-BMI was calculated. The degree of over nutrition was quantified using reference chart (Cole et al) on BMI for male/and female children of age 2–18 years.
Waist circumference was also considered for everyone: it was measured to the nearest 0,1 cm with a non-elastic tape while the participant was standing at the end of normal expiration and at the midpoint between the last rib and the crest of the ilium in a horizontal plane. Waist circumference has been quantified with reference charts for age and sex and z-waist circumference was calculated. Waist to height ratio was calculated for every patient.

Skin folds (biceps, triceps, subscapular and suprailiac) were measured in every child in triplicate, as proposed by Lohman using a Holtain Ltd skinfold caliper. Systolic and diastolic blood pressure were measured three times while the subjects were seated, and the measurements were averaged for the analysis.

For air-displacement plethysmography we used a BOD POD instrument (BOD POD ® Body Composition System, Life Measurement Instruments, Concord, CA, USA) to measure body volume and thoracic lung volume. The BOD POD software (version 1.69) calculated whole body density as body weight divided by body volume and fat mass percentage using Siri's quation.

Dual Energy X-ray Absorptiometry (DEXA) was performed using a Lunar DXP-IQ whole body absorptiometer (Lunar Corporation, Madison, WI, USA): patients laid down with arms and legs at their sides during the 15-min scan, with a radiation exposure < 7 μSv. Manufacturer's software version 4,6b was used, and daily quality-assurance tests were performed according to the manufacturer's directions.

Bioelectrical Inpedance Analysis (BIA) was measured using a tetrapolar, multifrequency BIA system (InBody 170, InBody Australia, Miami, Queensland, Australia) introducing an alternating current (20 and 100 Khz) at the base of the toes and fingers. Patients were standing with upper and lower arms slightly abduced for positioning of the electrodes. Impedance index was calculated as BH^2 divided by segmental impedance (m^2/ohm).

Ultrasound measurement was done on empty stomach in supine position and visceral fat was measured at the end of normal expiration with vertebral column positioned horizontal. The transducer was placed vertically and as lightly as possible to prevent compression of fat layers. Each measurement was performed 3 times and the average was considered for calculations. According to Armellini et al., we considered measurements of abdominal subcutaneous skin-hepatic thickness (pre-hepatic AT), abdominal subcutaneous skin-muscle thickness (umbilical AT), intra-abdominal muscle-aorta thickness (Aortic AT).

Blood samples were collected in the morning after overnight fasting for all the subjects included in the study. Metabolic biomarkers included total cholesterol, high density lipoprotein cholesterol (HDL), low density lipoprotein cholesterol (LDL), triglycerides (TG), fasting blood glucose, insulin, renal function as creatinine and blood urea nitrate, hepatic function as glutamic pyruvic transaminase (GPT), glutamic oxaloacetic transaminase (GOT) and gamma glutamyl transpeptidase (GGT). GOT levels were considered to be normal until 35 IU/L, GPT level until 38 IU/L and GGT levels were considered to be normal until a value of 42 IU/L. Cardiometabolic risk factors were identified as follows: hyper-triglyceridemia was defined as fasting triglycerides ≥150 mg/dl; low HDL-cholesterol as fasting HDL <40 mg/dl; hyperglycemia as fasting glucose ≥100 mg/dl. Homeostasis model assessment of insulin resistance (HOMA-IR) was calculated for each patient using the formula fasting insulin (mIU/l) x fasting glucose (mg/dl)/405. Each patient was evaluated according to the criteria of the metabolic syndrome; metabolic syndrome was considered according to the International Diabetes Federation (IDF) criteria:

- in children aged between 6 and 10 years, the metabolic syndrome is an entity that cannot be diagnosed, but the presence of a waist circumference higher than the 90th percentile and familiarity for metabolic syndrome, diabetes mellitus, or dyslipidemia are warning signs
- in children aged between 10 and 15 years, metabolic syndrome is defined by the presence of a waist circumference greater than the 90th percentile and the presence of at least two of the following signs: hypertriglyceridemia (≥150 mg/dl), hyperglycemia (≥100 mg/dl), low HDL (<40 mg/dl) or high blood pressure (≥130 mmHg for the systolic and ≥85 mmHg for the diastolic)
- in subjects over 15 years, the metabolic syndrome is considered in the presence of waist circumference greater than the 90th percentile associated with the presence of at least 2 of the following signs: triglyceridemia ≥150 mg/dl or pharmacological treatment for hypertriglyceridemia, fasting glycemia ≥100 mg/dl or diabetes

mellitus type 2, HDL <40 mg/dl for males and <50 mg/dl for females or pharmacological treatment for low HDL, blood pressure ≥130 mmHg for the systolic and ≥85 mmHg for the diastolic or pharmacological treatment for arterial hypertension

Statistical analysis was conducted using SPSS-20; correlation co-efficient and its significance were assessed between anthropometric measurements (z-height, z-weight, z-BMI, z-waist circumference and skin folds), visceral fat (prehepatic, umbilical) and blood values. Linear regression was used for those values which showed significant positive correlation. Logistic regression was used to estimate odds ratio between VAT and metabolic derangements. P value <0.05 was considered significant.

3.2 RESULTS

The characteristics of our group of patients are presented in Table 1. The mean age of the sample was 14,6 years (range 11–17 years), the same for the two subgroups females and males (p 0,807). The mean BMI z-score was 1,38 (± 0,7) for the whole cohort, 1,70 (± 0,77) and 1,25 (± 0,63) for males and females, respectively (p 0,007). Prevalence of overweight and obesity (BMI z-score higher than 1,036 and 1,645, referred to 85° percentile and 95° percentile of Center for Disease Control BMI charts) was 32,5% and 38,5% for the total of the patients respectively; for what concerns males and females, we found 16% (4/25) of overweight and 68% (17/25) of obese in the males, and 39,6% (23/58) and 25,8% (15/58) for the females, as reported in the table 3, resulting the prevalence of obesity to be significantly higher in our cohort in males than females ($p < 0,05$).

In addition, the overall sample had a mean value of 26,7 ± 9,4 kg for total body fat measured with DEXA, 2,84 ± 1,13 cm for subcutaneous abdominal fat and 3,32 ± 1,35 cm for visceral fat measured with ultrasound. Student's t tests revealed that boys had significantly higher visceral fat (4,18 ± 1,70 cm vs. 2,95 ± 0,96 cm; p ,< 0.05), including VAT/SAT ratio. After matching patients from the two genders in order to eliminate initial differences of z-BMI, the group of males still presents significantly higher mean VAT compared to females (4,18 cm vs, 3,30 cm respectively, $p < 0,05$). Total fat free mass and trunk fat/limbs fat ratio were also significantly higher in males than in females. No other significant differences were found between gender of patients. All results regarding anthropometric measurements by gender are shown in table 1.

In the table 2 we show the distribution of biochemical markers in our population of patients, divided for sex. Mean values of lipid profile, glycemia, insulin and hepatic function are under upper limits, but mean values of HOMA index are high for the general population and for both males and females group.

Statistically significant correlations between males and females are also shown in table 2. The two genders have similar total cholesterol and LDL profile, but differences are evident for all the other parameters. Males have lower mean HDL profile (50 mg/dL ± 12,4 vs. 61 mg/dL ± 12,7) compared to females, and higher tryglicerides (105 mg/dL ± 143, 1 vs. 63 mg/dL ± 29,1), fasting glucose (93 mg/dL ± 6,1 vs. 86 mg/dL ± 6,6), insulin (17 mIU/l ± 12,2 vs. 11 mIU/l ± 6,1), insulin resistance intended as higher HOMA index (4,16 ± 3,08 vs. 2,51 ± 1,42), GOT (23 IU/l ± 5,0 vs. 18 IU/l ± 5,2), GPT (24 IU/l ± 8,8 vs. 16 IU/l ± 6,8) and γGT (21,4 IU/l ± 11,1 vs. 12 IU/l ± 6,6).

Table 3 presents the proportion of participants with each metabolic risk factor; in the last line we reported the prevalence of metabolic syndrome in our sample of patients and its distribution among females and males, considering which subject had more than two risk factors associated to high waist circumference, as defined by IDF criteria for metabolic syndrome in evolutive age. The prevalence of metabolic syndrome among the two genders is significantly different: 25% of males (6/25) presented with the criteria for metabolic syndrome, while none of the females (0/58) could be included with the criteria of metabolic syndrome.

Bivariate analysis (Table 4) indicated that our anthropometric measurements of abdominal fat (superficial abdominal tissue, visceral abdominal tissue, VAT/SAT, fat percentage, total fat and trunk/limbs fat) were directly significantly associated with the following anthropometric measurements: BMI z-score and waist circumference z-score. In particular, in the linear regression model, VAT was significantly associated to all the other anthropometric measurements with a significant correlation, even if adjusted for age and sex and with physical activity level (table 5 to 7 and graphs 1 to 6).

As regards metabolic derangements, visceral abdominal fat, subcutaneous abdominal fat and their ratio VAT/SAT were correlated to blood values in a linear regression model adjusted for BMI z-score. VAT demonstrated to be superior than SAT and VAT/SAT in dectecting blood abnormalities, as it had significant correlation with fasting

glycemia, insulin, HOMA index and γGT, while VAT/SAT ratio only with insulin and HOMA, and SAT with GOT. None of the other blood values were positively correlated with visceral fat measurements. All the results are showed in table 8.

In order to see if visceral fat could be a major risk for metabolic derangements and for metabolic syndrome, we calculated odds ratio (OR) for VAT compared to MS and its criteria in evolutive age (high fasting glucose, low HDL cholesterol, high triglycerides and high arterial blood pressure) and other blood biomarkers such as liver enzymes. We didn't see any significant statistical correlation, excepted for the relationship between visceral fat and γGT, which showed a positive significant correlation with an odds ratio of 11,3. We also wanted to test if other indirect fat measurements could be a risk factor for metabolic derangements, but waist circumference didn't show any significant OR with the studied variables, while total fat measured with DEXA showed a significant correlation with γGT as well as found with VAT measured by ultrasonography. At last, we tried to calculate OR between VAT measure and the median of all blood biomarkers for our patients, in order to see if we could find an increased risk of higher blood values (but not considered pathological) for lipid status, liver enzymes and glucose metabolism. We saw VAT to have positive correlation with GPT higher than median ($p < 0,05$), with an OR of 1,6. Other matching didn't show any significant result. All the studies we performed to assess odds ratio are displayed in table 9, table 10, table 11 and table 12.

3.3 DISCUSSION

Obesity is known to be a major public health problem. Due to its currently increasing high prevalence and adverse effects on health, our life expectancy has been projected to decline for the first time since the last decades. This is in part due to obesity increasing significantly risk for cardio-metabolic diseases, such as cardiovascular disease and type-2 diabetes mellitus, which in turn are the major causes of morbidity and mortality in the industrialized world. This obesity-related risk for cardio vascular diseases is also a consequence of adipose tissue releasing a number of adipocytokines that, once released into the circulation, promote the development of metabolic derangements.

For many years, obesity has been assessed with body mass index which is an imprecise, and possibly misleading, metric of body fat; waist circumference and waist to height ratio are also easy to measure. BMI and waist circumference are influenced not only by fat mass but also by muscle mass and bone mass, among others. Furthermore, obesity-related risk for cardiovascular diseases increases not only with the quantity but also with a specific distribution of body fat: individuals who store body fat viscerally rather than elsewhere in the body (mostly subcutaneously) are at a greater risk for CVD. Several mechanistic pathways have been proposed to underlie the link between visceral fat and cardiovascular diseases: intra abdominal adipose tissue, as compared with subcutaneous adipose tissue, exhibits a more adverse secretory profile and higher lipid turnover. Further, visceral fat ~~but~~ drains directly to the portal circulation and liver, where it enhances dyslipidemia and insulin resistance, key mediators of the link between obesity and cardiometabolic diseases. The role of visceral fat involvement in metabolic derangements in adulthood has been largely investigated, but it's still under study for what concerns evolutive age. Despite the critical role of VF in metabolic syndrome pathogenesis and the recent emergence of MS in adolescence, only a few large-scale population-based studies quantified visceral fat directly. Magnetic Resonance Imaging and Computed Tomography scan are known to be

the most precise methodic to study body fat composition, but because of their costs and the problem of radiations, other accurate fat mass evaluations need to be found. In most population-based studies, abdominal obesity is measured indirectly with WC.
First aim of our study was to evaluate visceral fat with ultrasonography and see its correlation with other fat tissue assessment during evolutive age.
Our cohort of patients showed a significant difference of VAT between the two sexes, even after matching subjects for BMI, like Kotani et Al. and Goran et Al. already postulated in 1994 and 1995: sexual dimorphism of visceral fat supports that there is a definite gender difference in the age-related changes in whole-body fat distribution, especially in the abdominal fat tissues; sex differences in intraabdominal adipose tissue begin to emerge during pubertal development, with boys having more VAT than girls.
The present study revealed a significant correlation between visceral fat and all the other anthropometric measures; regression analysis showed that among the measurements used in this study, the best predictor of visceral fat is waist to height ratio. Although the best predictor of ultrasonography measured visceral fat, as per this study, is waist to height, as already stated before the predictive power of WC is low as WC does not distinguish visceral adiposity from the amount of subcutaneous abdominal fat, and thus, it cannot reflect fat redistribution and the coincidence rate of WC with VFA changes with the increase of age; it probably should therefore be considered a marker of total adiposity rather than VAT. With the data in our possess so far, we want to futher investigate this problem, in order to establish if assessment of visceral fat in children should be better done by an imaging modality and not indirectly from anthropometric measurements.
Previous studies have revealed that visceral adipose tissue has high activities of both lipogenesis and lipolysis, and its accumulation induces a high content of free fatty acids in portal circulation which goes into the liver directly. Excess free fatty acid may cause the enhancement of lipid synthesis and gluconeogenesis as well as insulin resistance, resulting in

hyperlipidemia, glucose intolerance, hypertension, and atherosclerosis. Thus, excess visceral adipose tissue is believed to be an important contributor to the development of cardiovascular diseases. In the present study, VFA was observed to be closely associated with glucose metabolic risk factors including high fasting glucose and insulin resistance. This finding confirms that visceral adipose tissue is associated with glucose metabolism in evolutive age as well as in adult age, as already postulated.

As regards hepatic function, no significant correlations were found between visceral fat and transaminase; nevertheless a strong correlation was found with γGT, demonstrating visceral fat to be an important predictor for hepatic functioning derangements.

The association between visceral adipose tissue and lipid metabolism biomarkers is not so evident in our study. After adjustment for age, sex and BMI, we didn't find any significant correlation between intra abdominal adipose tissue and lipid status. A high triglycerides level has been shown to be a risk factor for cardiovascular diseases, while HDL is inversely correlated with the risk of CVD and a low HDL level is one of the hallmarks of the metabolic syndrome, but none of this derangements where significantly correlated with the visceral fat accumulation in our cohort of patients.

Last aim of our study was to see if the increment of visceral fat could be a major predictor of the risk for metabolic derangements. The results we had from odds ratio statistical analysis show that no significant relationship could be found between VAT and the presence of metabolic syndrome or its diagnostic criteria such as high triglycerides, high fasting glucose, low HDL, high blood pressure; a good correlation, even if not significant, was found with insulin resistance, suggesting once again visceral fat deposit to be involved in glucose derangements and in diabetes mellitus type 2 onset. The only significant correlation was found with VF to be a major risk to have high γGT, showing an odds ratio of 11,3. To better investigate the role of visceral fat accumulation and metabolic derangements, we tried to calculate odds ratio between VAT and the risk of being above median value for each blood test: once

again, significant correlation was found between US visceral fat and GPT higher than the median value, showing how VAT accumulation is involved in liver functioning. This finding is very important as it shows how the VAT can be a method that reveals on a very early stage metabolic risk factors, before they reach values above the threshold. Limitations in this study could be that we only performed cross sectional study, and it doesn't allow causality interferences, but only associations; moreover the population we studied is not representative for the general population. At last, ultrasonography is not the gold standard although it is validated. On the other hand, our points of strength are that direct measurements of fat amount with gold standard method and direct measure of fat distribution although not gold standard showed important elements in favor of the use of the U.S. in the normal clinical practice.

The results we have presented among children and adolescent are very similar to those presented for adults so far, showing probably similar pathways in the visceral fat accumulation and in its metabolic significance. Further studies will be held to better investigate the importance of visceral fat accumulation in evolutive age as a predictor for metabolic derangements.

3.4 REFERENCES

- Agostoni et Al., 2011, Role of dietary factors and food habits in the development of childhood obesity: a commentary by the ESPGHAN Committee on nutrition, JPGN 52: 662-669
- Anis AH et Al., 2010, Obesity and overweight in Canada: an updated cost-of-illness study, Obes Rev 11: 31-40
- Armellini et Al., 1990, The contribution of sonography to the measurement of intra-abdominal fat, J Clin Ultrasound 18: 563-567
- Bacha F et Al., 2003, Obesity, regional fat distribution, and syndrome X in obese black versus white adolescents: race differential in diabetogenic and atherogenic risk factors, J Clin Endocrinol Metab 88: 2534 - 2540
- Baldisserotto et Al., 2013, Subcutaneous fat: a better marker than visceral fat for insulin resistance in obese adolescents, e-SPEN Journal 8: 251-255
- Barbeau P et Al., 2007, Ten months of exercise improves general and visceral adiposity, bone, and fitness in black girls, Obesity 15: 2077-2085
- Bertoli S et Al., 2008, Evaluation of air displacement plethysmography and bioelectrical impedance vs dual-energy X-ray absorptiometry for the assessment of fat-free mass in elderly subjects, Eur J Clin Nutr 62 (11): 1282-1286
- Bjorntorp P, 1992, Abdominal fat distribution and the metabolic syndrome, J Cardiovasc Pharmacol, 20(suppl 8): 526–528
- Brambilla P et Al., 1999, Persisting obesity starting before puberty is associated with stable intraabdominal fat during adolescence, Int J Obes Relat Metab Disord 23: 299–303
- Buyken AE et Al., 2008, Relation of dietary glycemic index, glycemic load, added sugar intake, or fiber intake to the development of body composition between ages 2 and 7 y, Am J Clin Nutr 88:755–62
- Caprio S et Al., 1996, Fat distribution and cardiovascular risk factors in obese adolescent girls: importance of the intraabdominal fat depot, Am J Clin Nutr 64: 12-17

- Cole TJ et Al., 2000, Establishing a standard definition for child overweight and obesity worldwide: international survey, BMJ 320: 1240-1243
- de Ridder et Al., 1992, Body fat distribution in pubertal girls quantified by Magneti Resonance Imaging, Int J Obes Relat Metab Disor 16: 443-449
- Demerath EWet Al., 2008 Visceral adiposity and its anatomical distribution as predictors of the metabolic syndrome and cardiometabolic risk factor levels: Am J Clin Nutr 88: 1263–1271.
- Després JP et Al., 1988, Relationships between body fatness, adipose tissue distribution and blood pressure in men and women. J Clin Epidemiol 41: 889–897
- Després JP et Al., 1985, Evidence for a regional component of body fatness in the association with serum lipids in men and women, Metabolism 34: 967–973
- Ellis KJ, 1997, Visceral fat mass in childhood: a potential early marker for increased risk of cardiovascular disease, Am J Clin Nutr 65:1887-1888
- Fox CS et Al., 2007, Abdominal visceral and subcutaneous adipose tissue compartments: association with metabolic risk factors in the Framingham Heart Study, Circulation 116: 39–48
- Fox K et Al., 1993, Abdominal fat deposition in 11-year-old children, Int J Obes Relat Metab Disord 17: 11–16
- Goodman E et Al., 2007, Instability in the diagnosis of metabolic syndrome in adolescents, Circulation 115: 2316-2322
- Goran MI, Gower BA, 1999, Relation between visceral fat and disease risk in children and adolescents, Am J Clin Nutr 70(suppl): 149S–156S
- Goran MI et Al., 1995, Intra-abdominal adipose tissue in young children, Int J Obes Relat Metab Disord 19: 279–283
- Gortmaker SL et Al., 2011, Changing the future of obesity: science, policy and action, the Lancet 378: 838-847

- Hunsenberger M et Al., 2013, Infant feeding practices and prevalence of obesity in eight European countries - the IDEFICS study, Public Health Nutr 16 (2): 219-227
- Hwang MJ et Al., 2008, How useful is waist circumference for assessment of abdominal obesity in Korean pre-menopausal women during weight loss?, Asia Pac J Clin Nutr 17: 229
- Iwata F et Al., 1995, Intra-abdominal fat in obese children, Acta Paediatr Jpn 37: 617-620
- Kahn CR, 1994, Insulin action, diabetogenes, and the cause of type II diabetes, Diabetes 43: 1066–1084
- Katzmarzyk PT et Al., 2012, Adiposity in children and adolescents: correlates and clinical consequences of fat stored in specific body depots, Pediatr Obesity 7: e42-e61
- Kim Y, Lee S, 2009, Physical activity and abdominal obesity in youth, Appl Physiol Nutr Metab 34: 571-581
- Kim Y, Lee S, 2009, Physical activity and abdominal obesity in youth, Appl Physiol Nutr Metab 34: 571-581
- Kissebah AH et Al., 1982, Relation of body fat distribution to metabolic complications of obesity, J Clin Endocrinol Metab 54: 254–260
- Kotani K et Al., 1994, Sexual dimorphism of age-related changes in whole-body fat distribution in the obese, Int J Obes Relat Metab Disord 18: 207-212
- Kytnarová et Al., 2004, Ultrasound measurement of visceral fat in obese children, Cas Lek Cesk 143: 166-169
- Lara J et Al., 2013, Accuracy of three novel predictive methods for measurements of fat mass in healthy overweight older subjects, Aging Clin Exp Res (Epub ahead of print)
- Lemieux S et Al., 1993, Sex differences in the relation of visceral adipose tissue accumulation to total body fatness, Am J Clin Nutr 58: 463–467
- Li X et Al., 2012, Visceral fat area, waist circumference and metabolic risk factors in abdominally obese Chinese adults, Biomed Environ Sci 25 (2); 141-148

- Lloyd T et Al., 1998, Body composition development of adolescent white females: the Penn State Young Women's Health Study, Arch Pediatr Adolesc Med 152: 998–1002
- Must A et Al., 1999, The disease burden associated with overweight and obesity, JAMA 282: 1523-1529
- Owens S et Al., 1999, Effect of physical training on total and visceral fat in obese children, Med Sci Sports Exerc 31: 143–148
- Pacifico L et Al., 2011, Menagement of metabolic syndrome in children and adolescents, Nutr Metab Cardiovascular Dis 21: 455-466
- Pausova Z, 2006, From big fat cells to high blood pressure: a pathway to obesity-associated hypertension, Curr Opin Nephrol Hypertens 15: 173-178
- Pinhas-Hamiel O et Al., 1996, Increased incidence of non-insulin-dependent diabetes mellitus among adolescents, J Pediatr 128: 608-615
- Pouliot MC et Al., 1994, Waist circumference and abdominal sagittal diameter: Best simple anthropometric indexes of abdominal visceral adipose tissue accumulation and related cardiovascular risk in men and women, Am J Cardiol 73: 460-468
- Rankinen T et Al., 1999, The prediction of abdominal visceral fat level from body composition and anthropometry: ROC analysis, Int J Obes Relat Metab Disord 23:, 801
- Reinehr T, Andler W, 2004, Changes in the atherogenic risk factor profile according to degree of weight loss, Arch Dis Child 89: 419-422.
- Sankar J et Al., 2012, Visceral fat assessment in over nourished children by ultrasonography and its relation to anthropometry, Indian J Pediatr 79 (10): 1338-1341
- Schwartz RS et Al., 1991, The effect of intensive endurance exercise training on body fat distribution in young and older men, Metabolism 40: 545–551
- Sinha R et Al., 2002, Prevalence of impaired glucose tolerance among children and adolescents with marked obesity, N Engl J Med 346: 802-10

- Sorof JM et Al., 2004, Overweight, ethnicity, and the prevalence of hypertension in school-aged children, Pediatrics 113: 475-482
- Spinelli A et Al., 2012, Sistema di sorveglianza OKkio alla salute: risultati 2010, Rapporti ISTISAN 12/14
- Steinberger J et Al., 2009, Progress and challenges in metabolic syndrome in children and adolescents: a scientific statement from the American Heart Association Atherosclerosis, Hypertension, and Obesity in the Young Committee of the Council on Cardiovascular Disease in the Young; Council on Cardiovascular Nursing and Council on Nutrition, Physical Activity, and Metabolism, Circulation 119: 628-647
- Suliga E, 2009, Visceral adipose tissue in children and adolescents: a review, Nutr Res Rev, 22: 137-147
- Syme C et Al., 2008, Intra-abdominal adiposity and individual components of the metabolic syndrome in adolescence: sex differences and underlying mechanisms, Arch Pediatr Adolesc Med 162: 453-461
- Terry T et Al., 2001, Growth of Visceral Fat, Subcutaneous Abdominal Fat, and Total Body Fat in Children, Obes Res 9: 283-289
- Toselli S et Al., 2012, Prevalence and tracking of weight disorders in Italian primary school students: a three-year follow-up, Coll Antropol 1: 63-67
- Tracy RE et Al., 1995, Risk factors and atherosclerosis in youth autopsy findings of the Bogalusa Heart Study, Am J Med Sci 310: S37–41
- Treuth MS et Al., 1995, Strength training reduces intra-abdominal adipose tissue in older women, J Appl Physiol 78: 1425–1431
- Vague J et Al., 1988, Fat distribution, obesities and health: evolution of concepts. In: Bouchard C, Johnston FE, eds. Fat distribution during growth and later health outcome. New York: Alan R Liss, 9–41
- Vega GL et Al., 2006, Influence of body fat content and distribution on variation in metabolic risk, J Clin Endocrinol Metab 91: 4459–4466

- Wang Y, Lobstein T, 2006, Worldwide trends in childhood overweight and obesity, Int J Pediatr Obes 1:11-2
- Zimmet P et Al., 2007, The metabolic syndrome in children and adolescents e an IDF consensus report, Pediatr Diabetes 8: 299-306

4 TABLES AND GRAPHS

Table 1

	TOTAL (n 83)			MALES (n 25)			FEMALES (n 58)		
	MIN	MAX	MEAN (SD)	MIN	MAX	MEAN (SD)	MIN	MAX	MEAN (SD)
AGE (years)	11	17	14,64 (1,90)	11	17	14,56 (2,06)	11	17	14,67 (1,84)
Z-BMI	-0,87	1,81	1,38 (0,70)	-0,87	2,81	1,70 (0,77)	-0,52	2,40	1,25** (0,63)
Z-WC	-0,34	7,74	3,40 (1,68)	-0,34	7,74	3,52 (1,99)	-0,03	6,58	3,34 (1,55)
SAT (cm)	0,33	5,79	2,84 (1,13)	0,33	5,79	3,01 (1,32)	0,44	5,41	2,76 (1,04)
VAT (cm)	1,18	8,57	3,32 (1,35)	1,47	8,57	4,18 (1,70)	1,18	5,56	2,95** (0,96)
VAT/SAT	0,40	7,06	1,39 (0,97)	0,62	7,06	1,77 (1,41)	0,40	4,36	1,23 (0,65)
FFM (Kg)	28,1	86,4	46,7 (11,1)	28,1	86,4	54,91 (15,38)	28,2	55,4	43,25 (6,01)
FAT %	9,4	54,1	35,8 (8,2)	9,4	48,9	33,38 (10,54)	16,9	54,1	36,89** (6,88)
TOT FAT (g)	7366	61951	26764 (9467)	7366	61951	28851 (13451)	9771	41670	25865** (7073)
TRUNK/LIMB	0,51	1,43	0,86 (0,18)	0,62	1,43	0,93 (0,2)	0,51	1,25	0,83 (0,16)

Abbreviation: BMI: body mass index; WC: waist circumference; VAT: visceral abdominal tissue; SAT: superficial abdominal tissue; FFM: fat free mass measured with BODPOD, TOT FAT: total fat measured with DEXA; TRUNK/LIMB: trunk fat/limbs fat ratio; **: significance $p < 0,05$

Table 2

	TOTAL (n 83)			MALES (n 25)			FEMALES (n 58)		
	MIN	MAX	MEAN (SD)	MIN	MAX	MEAN (SD)	MIN	MAX	MEAN (SD)
Chol. (mg/dL)	111	322	170 (43,4)	111	322	173 (43,4)	112	237	168 (27,8)
LDL (mg/dL)	48	160	96 (28,3)	54	160	103 (28,3)	48	150	93 (23,0)
HDL (mg/dL)	32	97	58 (12,4)	32	82	50 (12,4)	41	97	61** (12,7)
Triglyc (mg/dL)	23	759	75 (143,1)	23	759	105 (143,1)	29	211	62** (29,1)
Glycemia (mg/dL)	70	106	88 (6,1)	81	106	93 (6,1)	70	106	86** (6,6)
Insulin (mIU/L)	2,5	68,1	13 (12,2)	7,2	68,1	17 (12,2)	2,5	36,9	11** (6,1)
HOMA	,51	16,99	3,00 (3,08)	1,71	16,99	4,16 (3,08)	,51	7,92	2,51** (1,42)
GOT (IU/L)	11	34	19 (5,0)	14	34	23 (5,0)	11	34	18** (5,2)
GPT (IU/L)	9	48	18 (8,8)	13	42	24 (8,8)	9	48	16** (6,8)
γGT (IU/L)	4,0	49,4	15 (11,1)	10,5	49,4	21,4 (11,1)	4,0	48,9	12** (6,6)

Abbreviation: Chol: total cholesterol; LDL: low density lipoprotein; HDL high density lipoprotein; Triglyc: triglycerides; HOMA: Homeostasis Model for Assessment of Insulin Resistance; GOT: glutamic oxaloacetic transaminase; GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; **: significance $p < 0,05$

Table 3

	TOTAL	MALES	FEMALES
Overweight	27 (32,5%)	4 (16,0%)	23 (39,6%)
Obese	32 (38,5%)	17 (68,0%)	15 (25,8%)**
Hyperglycemia	5 (6,0%)	4 (16,0%)	1 (1,7%)**
Hypertriglycerides	4 (4,8%)	3 (12,0%)	1 (1,7%)**
Low HDL	7 (8,4%)	7 (28,0%)	0 (0%)**
High blood pressure	11 (13,2%)	8 (32,0%)	3 (5,1%)**
Metabolic syndrome	5 (6,0%)	6 (24,0%)	0 (0%)**
Insulin resistance	40 (48,1%)	17 (68,0%)	23 (39,6%)**

Abbreviation: HDL: high density lipoprotein; **: significance $p < 0,05$

Table 4

		Z-BMI	Z-WC	SAT	VAT	VAT/SAT	FAT %	TOT FAT	T/L
Z-BMI	R	1	0,807	0,711	0,518	-0,427	0,596	0,762	0,460
	SIG		#	#	#	#	#	#	#
Z-WC	R	0,807	1	0,800	0,471	-0,434	0,610	0,807	0,571
	SIG	#		#	#	#	#	#	#
SAT (cm)	R	0,711	0,800	1	0,396	-0,892	0,598	0,745	0,645
	SIG	#	#		#	#	#	#	#
VAT (cm)	R	0,518	0,471	0,396	1	-0,212	0,329	0,514	0,322
	SIG	#	#	#		#	#	#	#
VAT/SAT	R	-0,427	-0,434	-0,592	0,212	1	-0,431	-0,402	-0,324
	SIG	#	#	#	#		#	#	#
FAT %	R	0,596	0.610	0,598	0,329	-0,431	1	0,698	0,248
	SIG	#	#	#	#	#		#	#
TOT FAT (g)	R	0,762	0,823	0,745	0,514	-0,402	0,698	1	0,477
	SIG	#	#	#	#	#	#		#
T/L FAT	R	0,490	0,571	0,645	0,322	-0,324	0,248	0,477	1
	SIG	#	#	#	#	#	#	#	

Abbreviation: Z-BMI: body mass index z-score; Z-WC: waist circumference z-score; SAT: superficial asipose tissue; VAT: visceral adipose tissue; VAT/SAT: visceral adipose tissue/superficial adipose tissue ratio; FAT%: total fat percentage measured with BOD POD; TOT FAT: total fat mass measured with DEXA; T/L FAT: trumk fat/limb fat ratio; R: Pearson's correlation; SIG: significance (#: $p < 0,05$)

Table 5

	VAT				VAT (sport)			
	Coeff.	Stand.Er.	T	p	Coeff.	Stand.Er.	T	p
Z-WEIGHT	0,323	0,156	3,287	0,002**	0,313	0,155	3,211	0,002**
Z-BMI	0,425	0,188	4,320	0,000**	0,410	0,188	4,166	0,000**
Z-WC	0,444	0,071	5,023	0,000**	0,430	0,071	4,810	0,000**
WAIST/HEIGHT	0,542	1,986	6,132	0,000**	0,528	2,007	5,910	0,000**
BODY FAT %	0,419	0,015	4,515	0,000**	0,404	0,015	4,351	0,000**
FAT FREE MASS	0,101	0,017	0,740	0,461	0,127	0,016	0,935	0,353
TOTAL FAT	0,510	0,038	5,847	0,000**	0496	0,045	5,630	0,000**
TRUNK/LIMB	0,252	0,742	2,477	0,015**	0,241	0,739	2,375	0,020**

Abbreviation: Coeff; coefficient; Stand. Er.: standard error; VAT: visceral adipose tissue; VAT (sport): visceral adipose tissue regression corrected for physical activity; Z-WEIGHT: weight z-score; Z-BMI: BMI z-score; WAIST/HEIGHT: waist to height ratio; BODY FAT %: total fat percentage measured with BOD POD; FAT FREE MASS: total lean mass measured with BOD POD; TOTAL FAT: total fat mass measured with DEXA; TRUNK/LIMB: trunk fat/limbs fat ratio
SIG: **: significance $p < 0,05$

Table 6

	SAT				SAT (sport)			
	Coeff.	Stand.Er.	T	p	Coeff.	Stand.Er.	T	p
Z-WEIGHT	0,700	0,113	3,287	0,000**	0,691	0,112	8,199	0,000**
Z-BMI	0,805	0,129	10,035	0.000**	0,793	0,129	9,872	0,000**
Z-WC	0,805	0,045	12,108	0,000**	0,796	0,045	11,828	0,000**
WAIST/HEIGHT	0,829	1,386	11,295	0,000**	0,819	1,403	11,020	0,000**
BODY FAT %	0,658	0,012	7,468	0,000**	0,645	0,013	7,245	0,000**
FAT FREE MASS	0,401	0,015	2,778	0,007**	0,433	0,015	3,040	0,003**
TOTAL FAT	0,773	0,468	10,153	0,000**	0,762	0,438	9,896	0,000**
TRUNK/LIMB	0,672	0,546	7,528	0,000**	0,661	0,543	7,454	0,000**

Abbreviation: Coeff; coefficient; Stand. Er.: standard error; SAT: superficial asipose tissue; SAT (sport): superficial adipose tissue regression corrected for physical activity; Z-WEIGHT: weight z-score; Z-BMI: BMI z-score; WAIST/HEIGHT: waist to height ratio; BODY FAT %: total fat percentage measured with BOD POD; FAT FREE MASS: total lean mass measured with BOD POD; TOTAL FAT: total fat mass measured with DEXA; TRUNK/LIMB: trunk fat/limbs fat ratio
SIG: **: significance $p < 0,05$

Table 7

	VAT/SAT				VAT/SAT (sport)			
	Coeff.	**Stand.Er.**	**T**	**p**	**Coeff.**	**Stand.Er.**	**T**	**p**
Z-WEIGHT	-0,559	0,106	-6,013	0,000**	-0,563	0,106	-6,036	0,000**
Z-BMI	-0,611	0,130	-6,437	0,000**	-0,623	0,131	-6,535	0,000**
Z-WC	-0,458	0,055	-4,775	0,000**	-0,473	0,056	-4,890	0,000**
WAIST/HEIGHT	-0,427	1,689	-4,090	0,000**	-0,443	1,709	-4,190	0,000**
BODY FAT %	-0,412	0,012	-4,072	0,000**	-0,422	0,012	-4,136	0,000**
FAT FREE MASS	-0,411	0,012	-2,962	0,004**	-0,411	0,012	-2,923	0,005**
TOTAL FAT	-0,453	0,578	-4,547	0,000**	-0,468	0,569	-4,656	0,000**
TRUNK/LIMB	-0,416	0,541	-4,037	0,000**	-0,421	0,544	-4,063	0,000**

Abbreviation: Coeff; coefficient; Stand. Er.: standard error; VAT/SAT: visceral adipose tissue/superficial adipose tissue ratio; VAT/SAT (sport): visceral adipose tissue/superficial adipose tissue ratio regression corrected for physical activity; Z-WEIGHT: weight z-score; Z-BMI: BMI z-score; WAIST/HEIGHT: waist to height ratio; BODY FAT %: total fat percentage measured with BOD POD; FAT FREE MASS: total lean mass measured with BOD POD; TOTAL FAT: total fat mass measured with DEXA; TRUNK/LIMB: trunk fat/limbs fat ratio SIG: **: significance $p < 0,05$

Table 8

	VAT				SAT				VAT/SAT			
	Coeff.	Stand.Er.	T	p	Coeff.	Stand.Er.	T	p	Coeff.	Stand.Er.	T	p
Chol. (mg/dL)	0,039	0,004	0,421	0,675	0,014	0,003	0,182	0,856	0,042	0,003	0,467	0,6422
LDL (mg/dL)	0,112	0,005	1,161	0,249	0,082	0,004	1,048	0,298	0,047	0,004	0,500	0,618
HDL (mg/dL)	-0,079	0,010	-,768	0,445	-0,095	0,007	-,940	0,258	0,004	0,007	0,042	0,697
Triglyc (mg/dL)	-0,006	0,002	-,060	0,953	-0,042	0,001	-,534	0,595	0,014	0,001	0,149	0,882
Glycemia (mg/dL)	0,206	0,018	2,126	0,037*	0,220	0,013	2,794	0,086	0,013	0,013	0,132	0,896
Insulin (mIU/L)	0,288	0,016	2,680	0,009*	0,019	0,012	0,211	0,832	0,235	0,012	2,243	0,028*
HOMA	0,272	0,066	2,555	0,013*	0,017	0,047	0,192	0,848	0,214	0,046	2,055	0,043*
GOT (IU/L)	0,024	0,025	0,235	0,815	0,178	0,016	2,182	0,032*	-0,001	0,017	-,012	0,991
GPT (IU/L)	0,124	0,017	1,159	0,250	0,015	0,012	0,177	0,860	0,195	0,012	1,927	0,058
γGT (IU/L)	0,324	0,015	3,262	0,002*	0,049	0,010	0,572	0,569	0,149	0,011	1,477	0,144

Abbreviation: Coeff; coefficient; Stand. Er.: standard error; Chol: total cholesterol; LDL: low density lipoprotein; HDL high density lipoprotein; Triglyc: triglycerides; HOMA: Homeostasis Model for Assessment of Insulin Resistance; GOT: glutamic oxaloacetic transaminase; GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; VAT: visceral adipose tissue; SAT: superficial asipose tissue; VAT/SAT: visceral adipose tissue/superficial adipose tissue ratio; SIG: *: significance $p < 0,05$

Table 9

	VAT	
	SIGNIFICANCE (p)	ODDS RATIO
METABOLIC SYNDROME	0,681	1,156
HYPERGLYCEMIA	0,503	1,267
INSULIN RESISTANCE	0,059	1,704
HYPERCHOLESTEROLEMIA	0,754	1,089
HYPERTRIGLYCERIDEMIA	0,058	2,164
HYPO HDL	0,919	1,034
HIGH GOT	-	-
HIGH GPT	0,197	1,802
HIGH γGT	0,009**	11,332
HIGH BP	0,832	0,945

Abbreviation: HDL high density lipoprotein; GOT: glutamic oxaloacetic transaminase (no values above limit); GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; BP: arterial blood pressure; VAT: visceral adipose tissue; **: significance $p < 0,05$

Table 10

	Z-WC	
	SIGNIFICANCE (p)	ODDS RATIO
METABOLIC SYNDROME	0,514	0,719
HYPERGLYCEMIA	0,988	0,993
INSULIN RESISTANCE	0,705	1,102
HYPERCHOLESTEROLEMIA	0,628	0,848
HYPERTRIGLYCERIDEMIA	0,482	0,699
HYPO HDL	0,308	0,625
HIGH GOT	-	-
HIGH GPT	0,666	0,773
HIGH γGT	0,302	1,682
HIGH BP	0,420	0,757

Abbreviation: Z-WC: waist circumference z-score; HDL high density lipoprotein; GOT: glutamic oxaloacetic transaminase (no values above limit); GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; BP: arterial blood pressure; VAT: visceral adipose tissue; **: significance $p < 0,05$

Table 11

	TOTAL FAT	
	SIGNIFICANCE (p)	ODDS RATIO
METABOLIC SYNDROME	0,209	0,987
HYPERGLYCEMIA	0,331	0,630
INSULIN RESISTANCE	0,481	1,231
HYPERCHOLESTEROLEMIA	0,764	0,493
HYPERTRIGLYCERIDEMIA	0,941	0,899
HYPO HDL	0,226	0,475
HIGH GOT	-	-
HIGH GPT	0,348	0,787
HIGH γGT	0,014**	4,743
HIGH BP	0,757	0,797

Abbreviation: TOTAL FAT: total fat mass measured with DEXA; HDL high density lipoprotein; GOT: glutamic oxaloacetic transaminase (no values above limit); GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; BP: arterial blood pressure; VAT: visceral adipose tissue; **: significance $p < 0,05$

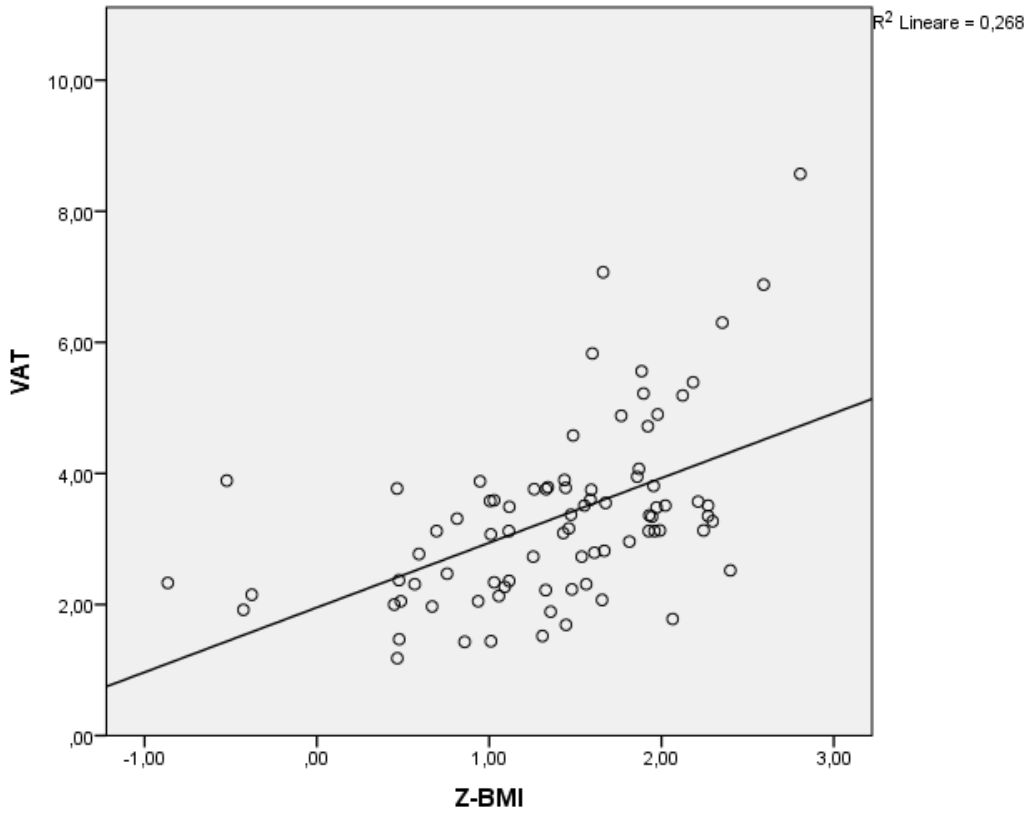
Table 12

	VISCERAL FAT	
	SIGNIFICANCE (p)	ODDS RATIO
HIGH GLYCEMIA	0,086	1,452
INSULIN RESISTANCE	0,079	1,647
HIGH CHOLESTEROL	0,255	1,257
HIGH LDL	0,248	1,282
HIGH TRIGLYCERIDES	0,697	1,079
HYPO HDL	0,084	1,453
HIGH GOT	1,839	1,040
HIGH GPT	0,038**	1,649
HIGH γGT	0,313	1,239

All the values tested are intended to be over (high) or under (low) the median value.

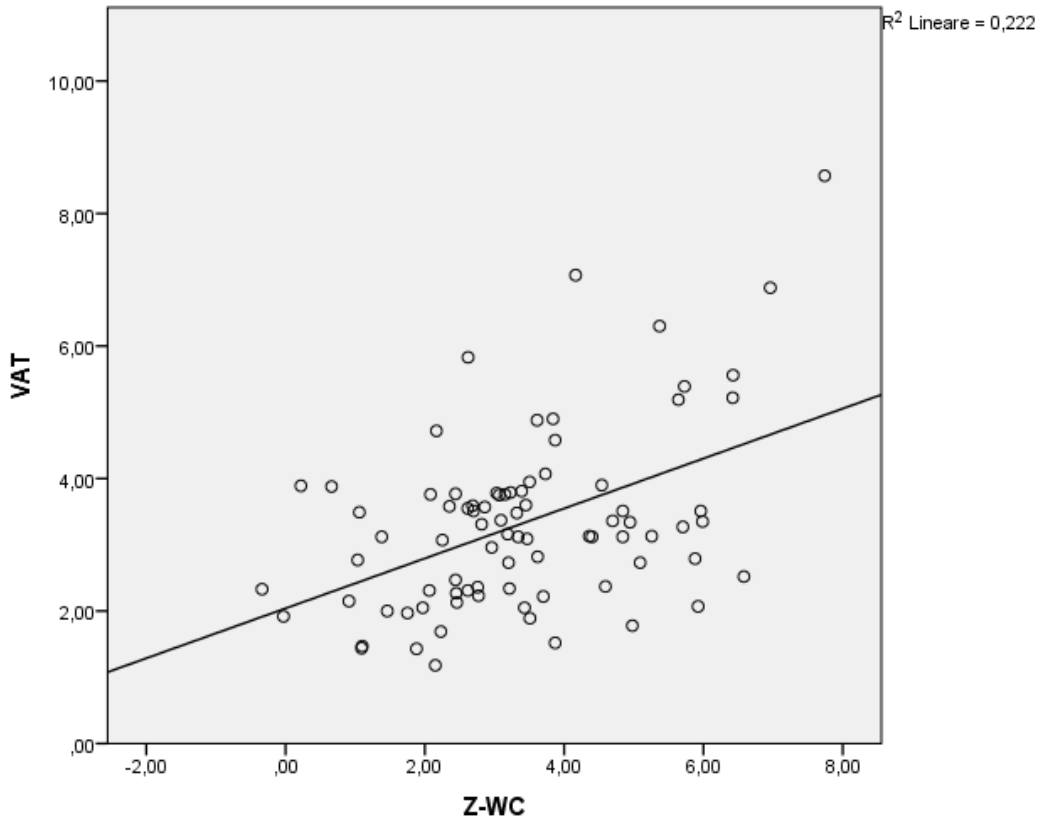
Abbreviation: LDL: low density lipoproteine; HDL high density lipoprotein; GOT: glutamic oxaloacetic transaminase (no values above limit); GPT: glutamic pyruvic transaminase; γGT: gamma glutamyl transpeptidase; VAT: visceral adipose tissue; **: significance $p < 0,05$

Graph 1



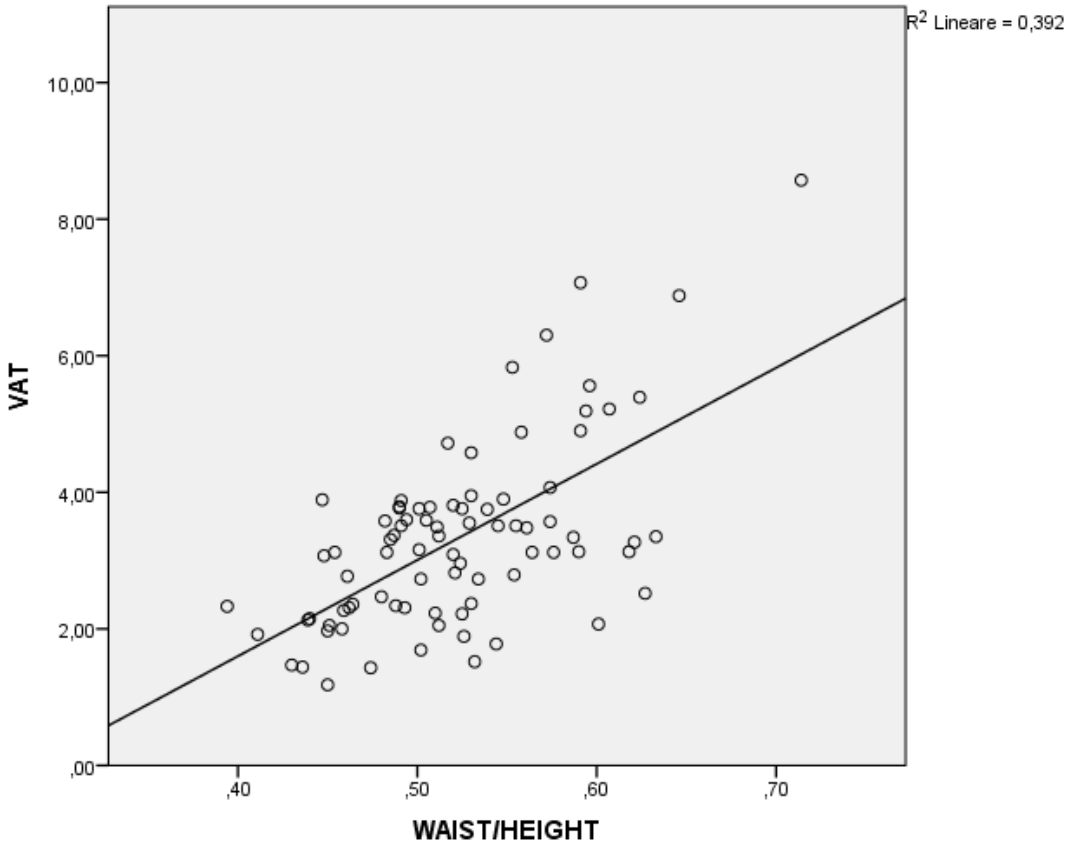
Abbreviations: VAT: visceral abdominal tissue; Z-BMI: body mass index z-score

Graph 2



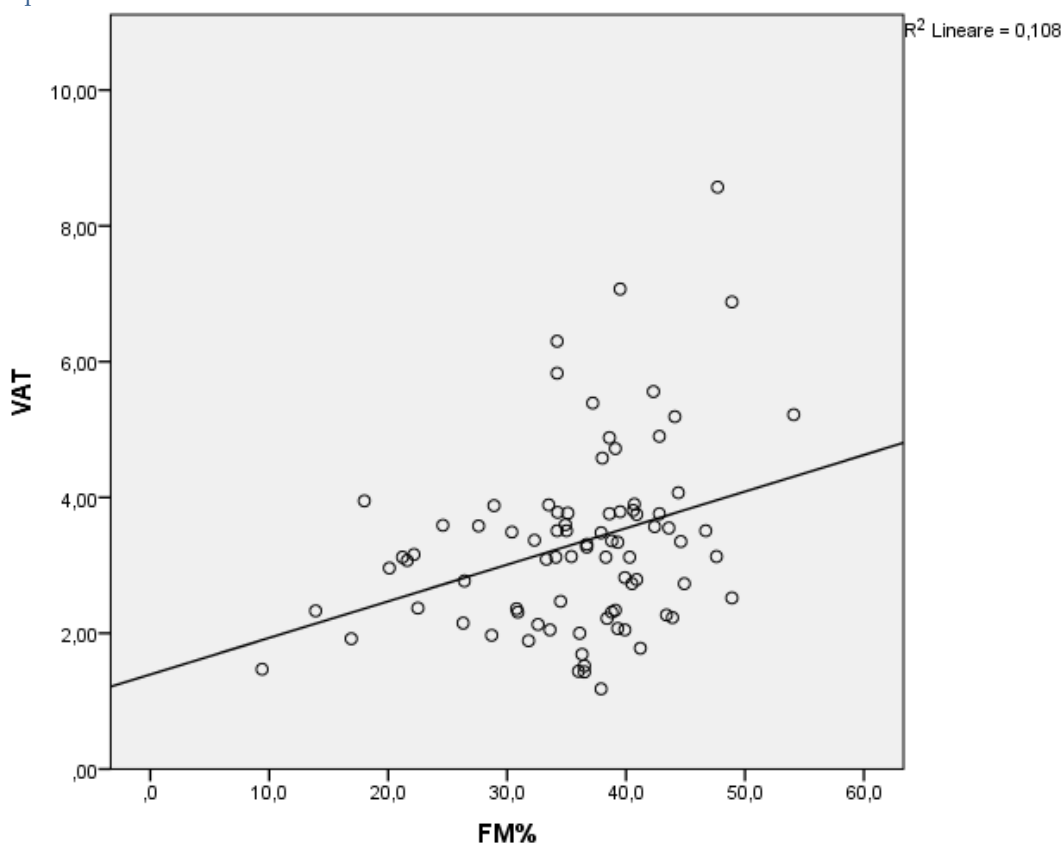
Abbreviations: VAT: visceral abdominal tissue; Z-WC: waist circumference z-score

Graph 3



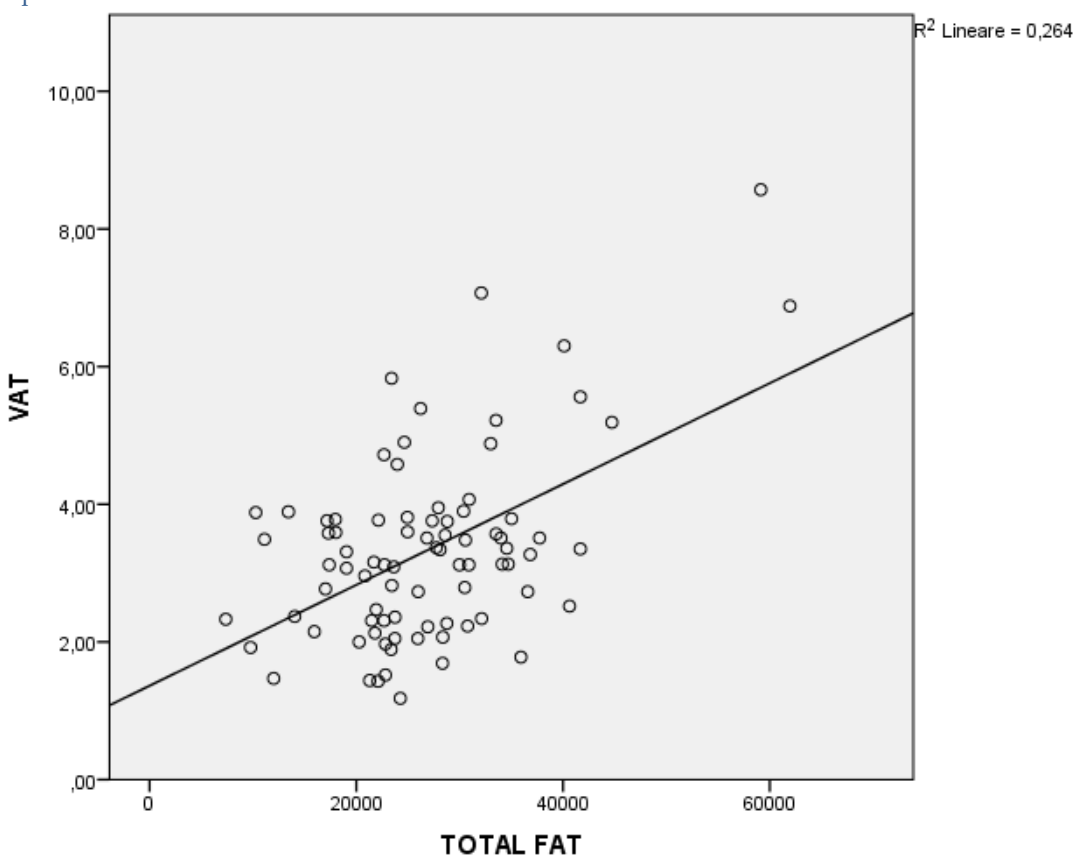
Abbreviations: VAT: visceral abdominal tissue; WAIST/HEIGHT: waist to height ratio

Graph 4



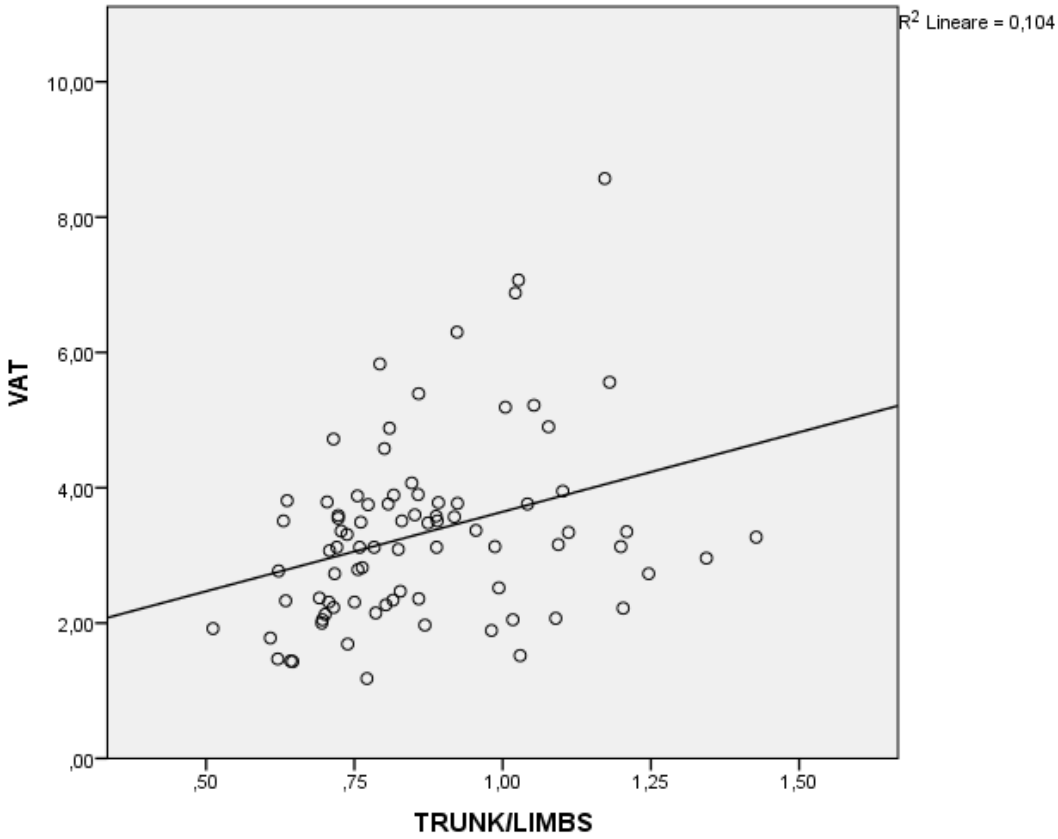
Abbreviations: VAT: visceral abdominal tissue; FM%: fat mass percentage measured with BOD POD

Graph 5



Abbreviations: VAT: visceral abdominal tissue; TOTAL FAT: total fat mass measured with DEXA

Graph 6



Abbreviations: VAT: visceral abdominal tissue; TRUNK/LIMBS: trunk fat/limbs fat measured with DEXA