

Dissertation



Medical University of Graz

**DETECTING BODY FAT – A WEIGHTY PROBLEM  
BMI versus SUBCUTANEOUS FAT PATTERNS**

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Ich erkläre ehrenwörtlich, dass ich die vorliegende Arbeit selbständig angefertigt und abgefasst, und jene Personen und Institutionen, die am Zustandekommen der Forschungsdaten beteiligt waren, namentlich genannt habe. Andere als die angegebenen Quellen habe ich nicht verwendet und die den benutzten Quellen wörtlich oder inhaltlich entnommenen Stellen habe ich als solche kenntlich gemacht. Die Arbeit an der Dissertation und daraus entstandener Publikationen wurde gemäß den Regeln der „Good Scientific Practice“ durchgeführt.

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Wien, am 03.03.2014

Unterschrift:

## Danksagungen/Expression of thanks

“Was du mir sagst, das vergesse ich. Was du mir zeigst, daran erinnere ich mich.

Was du mich tun lässt, das verstehe ich.“

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## **Zusammenfassung**

### **KÖRPERFETT DETEKTIEREN – EIN GEWICHTIGES PROBLEM BMI versus VERTEILUNG DER SUBKUTANEN FETTMASSE**

#### **Hintergrund**

Der Body Mass Index (BMI) ist eine weit verbreitete Methode zur Bestimmung von krankhaftem Übergewicht, jedoch misst er eher ein Zuviel an Gewicht, als einen Überschuss von Körperfett, in Relation zur Körpergröße. Das BMI-Klassifikations-System beruht auf Durchschnittswerten der allgemeinen Bevölkerung und ist nicht spezifisch genug für Subgruppen wie Athleten und jüngere Kollektive. Ein höherer Anteil an Muskelmasse bei Athleten und jüngeren Personen im Vergleich zur Allgemeinbevölkerung führt zu einer Fehlklassifizierung dieser Kollektive. Die Verteilung der subkutanen Fettmasse (SAT-Top) könnte einen effizienteren Zugang zur Evaluierung der Fettmasse bei Athleten und jüngeren Kollektiven darstellen.

#### **Ziele**

Die Ziele der Studie sind es, den Zusammenhang zwischen BMI und SAT-Top bei jungen Athleten und nicht-athletischen Kontrollen zu beschreiben, die Trennkraft von BMI und subkutanen Fettdicken an verschiedenen Messpunkten mittels ROC-Curve Analyse zu evaluieren und geeignete Lipometriemesspunkte zu eruieren, anhand derer es möglich ist die beiden Gruppen möglichst scharf zu trennen.

#### **Methoden**

Die Messung der Verteilung der subkutanen Fettmasse (SAT) wurde mittels Lipometer an 15 anatomisch definierten Stellen des Körpers vom Nacken bis zum Unterschenkel bei 64 Männern und 42 Frauen durchgeführt, die in zwei gleiche Gruppen (Athleten/innen und Nicht-Athleten/innen) geteilt wurden. Die Trennkraft der einzelnen Messpunkte und des BMI wurde mittels ROC-Curve-Analyse ermittelt.

**Ergebnisse**

Bei Männern zeigten der Nacken (optimaler Trennwert: 2.3 mm) und der Rumpf (optimaler Trennwert: 15.5 mm) die stärkste Trennkraft. 90.6% (58 von 64) der Studienteilnehmer wurden korrekt als Athleten oder Nicht-Athleten klassifiziert. Die Trennkraft der BMI-Werte erwies sich zwischen den Gruppen als nicht signifikant. Bei Frauen zeigten der obere Rücken (optimaler Trennwert: 3.3 mm) und die Arme (optimaler Trennwert: 15.9 mm) mit 88.1% (37 von 42) richtig klassifizierten Studienteilnehmerinnen die stärkste Trennkraft. Die Verwendung des BMI zur Trennung von Athletinnen und Nicht-Athletinnen führte lediglich zu 52.4% (22 von 42) korrekt klassifizierten Studienteilnehmerinnen.

**Zusammenfassung**

Die Resultate zeigen, dass die Verteilung der subkutanen Fettmasse im Vergleich zu BMI-Werten eine akkuratere Methode zur Unterscheidung zwischen Athleten und Nicht-Athleten darstellt. Besonders der Nacken und der Rumpf bei Männern und der obere Rücken und die Arme bei Frauen zeichnen sich als adäquate Trennpunkt zur Unterscheidung zwischen Athleten und Nicht-Athleten ab.

**Schlüsselworte**

Body Mass Index, Lipometer, junge Athleten, subkutanes Fett, ROC-Curve, Adipositas

## **Abstract**

### **DETECTING BODY FAT – A WEIGHTY PROBLEM BMI versus SUBCUTANEOUSE FAT PATTERNS**

#### **Background**

Although the body mass index (BMI,  $\text{kg/m}^2$ ) is widely used as a surrogate measure of adiposity, it is a measure of excess weight, rather than excess body fat, relative to height. The BMI classification system is derived from cut points obtained from the general adult population and is not specific for subgroups such as athletes and young adults. The use of subcutaneous adipose tissue topography (SAT-Top) seems to be a more effective access for such cases.

#### **Objective**

We aimed to describe the relationship between BMI and the subcutaneous adipose tissue topography within young athletes and non-athletic controls, to comparatively evaluate the diagnostic powers of BMI and subcutaneous adipose tissue thicknesses at different body sites, and to explore appropriate subcutaneous adipose tissue measuring points and cutoffs to discriminate between athletes and non-athletic controls.

#### **Methods**

Measurements were determined in 64 males and 42 females, who were subsequently separated into two even groups (athletes and non-athletes). The optical device Lipometer was applied at standardised body sites to measure the thickness of subcutaneous adipose tissue layers. To calculate the power of the different body sites and the BMI to discriminate between athletes and non-athletes, receiver operating characteristic curve analysis was performed.

#### **Results**

In men, the neck (optimal cutoff value 2.3 mm) and trunk (optimal cutoff value 15.5 mm) provided the strongest discrimination power with 90.6% (58 of 64) of the subjects being correctly classified into athletes or non-athletes. Discrimination

power of the BMI values was not significant. In women, the upper back (optimal cutoff value 3.3 mm) and arms (optimal cutoff value 15.9 mm) provided the strongest discrimination power with 88.1% (37 of 42) being correctly classified. When using BMI to discriminate between athletes and non-athletes only 52.4% (22 of 42) were correctly classified.

### **Conclusion**

These results suggest that compared to BMI levels, subcutaneous fat patterns are a more accurate way of discriminating between athletes and non-athletes. In particular the neck and the trunk compartment in men and the upper back and arms compartment in women, were the best sites discriminate between young athletes and non-athletes on the basis of their fat patterns.

### **Key Words**

Lipometer, Young Athletes, Subcutaneous Fat, ROC Curve, Body Mass Index, Obesity

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## Glossary and acronyms

The following abbreviations are used in this dissertation:

AACE	American Association of Clinical Endocrinology
ACE	American College of Endocrinology
ACSM	American College of Sports Medicine
ADP	Air displacement plethysmography
AI	Area index
AT	Adipose tissue
BCM	Body Cell Mass
BIA	Bioelectric impedance analysis
BM	Body Mass
BMI	Body Mass Index (kg/m <sup>2</sup> )
BV	Body Volume
CHO	Carbohydrates
CI	Confidential interval
cm	Centimetres
CT	Computed Tomography
CV	Coefficients of variation
CVD	Cardio vascular disease
D <sub>b</sub>	Density of the human body
D(E)XA	Dual energy X-ray absorptiometry
D <sub>w</sub>	Density of water
ECF	Extracellular fluid
ECM	Extracellular mass
ECS	Extracellular solids
ECW	Extracellular water
e.g.	For example
FM	Fat mass
FFM	Fat free mass
HD	Hydrometry
HR	Hazard ratio

IAAS	International anthropometry accreditation scheme
ICW	Intracellular water
ISAK	International Society for the Advancement of Kinanthropometry
kg	Kilogram
kHz	Kilohertz
l	Liters
L	Length
LBM	Lean body mass
m	Meter
M	Mineral
MA	Mass
mm	Millimeter
max	Maximum
min	Minimum
Mo	Bone mineral
Ms	Soft tissue mineral
MRI	Magnetic resonance imaging
NIH	National Institutes of Health
n.s.	Not significant
p	Significance level
P	Pressure
r	Correlation coefficient
R	Resistance
ROC	Receiver operating characteristic
SAT	Subcutaneous adipose tissue
SAT-Top	Subcutaneous adipose tissue topography
SD	Standard deviation
SEE	Standard error of estimate
SM	Skeletal muscle
TBF	Total body fat absolute
TBF%	Total body fat percentage
TBW	Total body water
TBPro	Total body protein

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US	Ultrasound
UWW	Under water weighing
V	Volume
$W_a$	Weight in air
$W_w$	Weight in water
WHO	World Health Organization
WHR	Waist-hip-ratio
X	Mean value
$X_c$	Reactance
3D	Three-dimensional
Y	Year

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# 1 Introduction

Since James S. Garrow proposed the body mass index (BMI, kg/m<sup>2</sup>) as a measure of fatness in 1985 (1), its use in science and within clinical practice has risen exponentially over the years. The search in the database “Web of Science” for the search term “Body Mass Index” did result in 11008 hits for the publications year 2012 compared to 103 hits for publications in 1990 (search date: April 2013).

Especially in sports science the BMI and assessment of body fat to determine optimal body weight has increased (2).

The Body mass index (BMI) is a simple index of weight-for-height that is commonly used to classify overweight and obesity (3). Overweight and obesity are defined by the World Health Organization (WHO), as abnormal or excessive body fat accumulation in such an extent, that health may be adversely affected (4). In both athletic and non-athletic populations the estimates of body composition characteristics are used to identify health status (5).

Body weight and body composition are important performance factors in many sports (6). A centralized subcutaneous fat distribution has been associated with decreased aerobic capacity in men (7).

Nevertheless the use of body weight by itself and/or BMI has been criticized, particularly in athletic populations (8-10). Due to its simple, fast and ubiquitous practicability it is a well-known and widespread measure of human obesity. But how eligible is the BMI compared to other measures of body composition effectively?

## **1.1 History of Body Composition**

First body composition concepts can already be found among the Greeks 400 before Christ (11). Anthropometry [Greek anthropos (άνθρωπος - "man") and metron (μέτρον - "measure") = "measurement of man"] refers to the measurement of living human individuals for the purposes of understanding human physical variation (12).

The modern era of body composition research can be traced back about one century, where scientists, such as Justus von Liebig, prepared substantial groundwork for the field (13).

In 1921 J. Matiegka, a Czech anthropologist, reported the first anthropometric model to estimate total body muscle mass which was an important scientific advance. With the introduction of metabolic concepts and stable and radioactive labeled isotopes the golden era of body composition was entered (13).

A benchmark was set by the introduction of the underwater weighing method and two-compartment model in the early 1940s (13). It was Albert Behnke, an American physician, which introduced this more practical method to measure fat mass (FM) and fat-free mass (FFM) in humans. He is therefore known as the "modern-day father" of human body composition (13).

Important methods as the whole-body  $^{40}\text{K}$  counting (1961) and in vivo neutron activation analysis (1964), as well as the bioelectric impedance analysis (BIA) method (1962), were introduced in the 1960s (13).

The origins of the modern era of body composition can be traced back to the introduction of the dual-energy X-ray absorptiometry (DXA) in the early 1970s and the development of computer tomography (CT) and magnetic resonance imaging (MRI) within the next two decades (13).

## **1.2 History of BMI**

In 1832 the polymath Lambert Adolphe Jacques Quetelet (1796-1874), born in Gent (Belgium), described the Quetelet Index as weight in kilograms divided by the square of the height in meters. He invented the Quetelet Index in the course of developing "social physics". In his book "A treatise on man and the development of his faculties", he wrote „If we now compare fully developed and regularly built individuals so to assess the relationships that might exist between weight and height, we will find that the weights among developed subjects of different heights are approximately like the squares of the heights. It thus follows that transverse section including width and thickness is simply like a man's height“. [entire paragraph (14)]

These words prove that he found the formula, but he did not propose it as a measure of body size or body fatness. His intent was defining the characteristics of an average man and to adjust the distribution around the norm. From 1831–1832 he directed what has been esteemed the first cross sectional study about children and newborns based on height and weight, and extended it to a study about adults (1, 14).

After the 2<sup>nd</sup> World War when actuaries reported the increased mortality of their overweight policyholders, they saw need for a practical index of relative body weight and so the Quetelet Index was discovered again in the 1960s (14).

The fourth examination of the Framingham study (1954 to 1958) was one of the first studies to confirm the validity of the Quetelet Index. In 1972 Ancle Keys (1904-2004) confirmed the validity of the Quetelet Index and renamed it the Body Mass Index (14, 15). Keys intended to use the BMI in population studies and not for individual diagnosis (14-16).

In the early 20<sup>th</sup> century the term polysarcia was gradually replaced by obesity. It was James S. Garrow, a pioneer in body composition and obesity management, which proposed the Quetelet Index to become a measure of fatness in 1985 (1). Furthermore he proved that the BMI (Quetelet Index) correlates well with fat mass (1).

### **1.3 The use of BMI**

Today clinicians and researchers use the BMI, calculated as body mass in kilograms divided by height in meters squared (equation 1), to classify adult body weight.

$$BMI = \frac{MA [kg]}{L^2 [m^2]}$$

**Equation 1 BMI formula**

To simplify the assessment in practice BMI charts (figure 1) or BMI discs (figure 2) are used. The chart displays the BMI as a function of weight (vertical axis) and height (horizontal axis) using different colours for several BMI categories or contour lines for different values of BMI (17). For individuals under the age of 18, overweight is defined as a BMI over the 90<sup>th</sup> percentile and obesity over 97<sup>th</sup> percentile of age-specific BMI growth charts percentile of age-specific BMI growth charts (18).

Due to the fast, safe, simple and inexpensive handling the use of the BMI has become the most popular method to classify human weight (19).

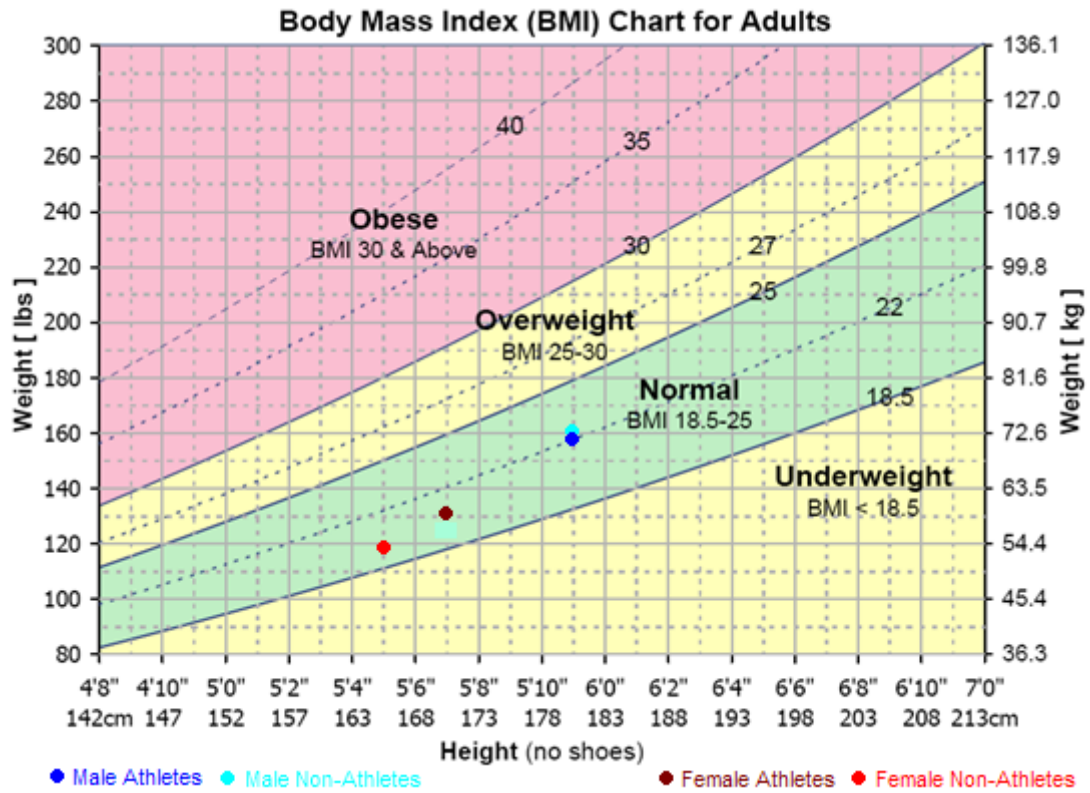


Figure 1 BMI chart for adults with BMI-study data from table 4 and 5 in comparison athletes vs. non-athletes [adapted from (20)]

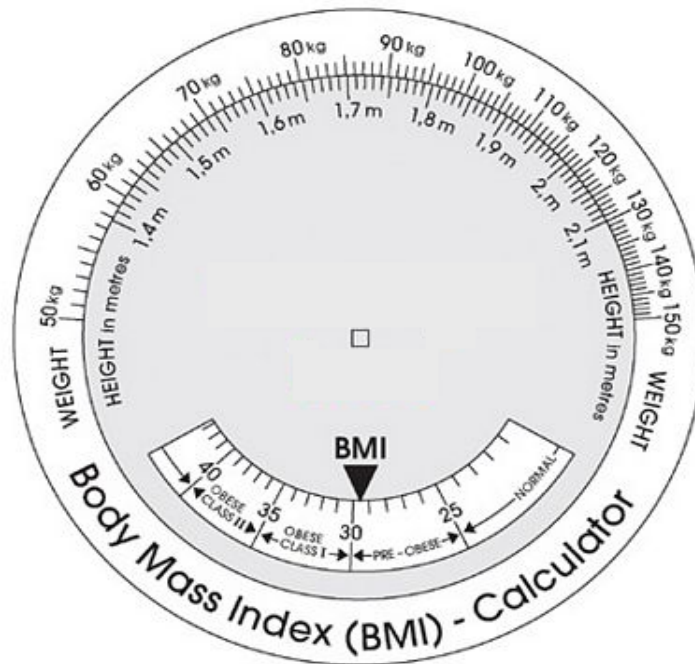


Figure 2 BMI disc for adults [adapted from (21)]

## **1.4 BMI Classification**

The first health-related BMI classification system derives from Garrow et al. (22) and was adopted in a modified form by the WHO (23) as shown in table 1.

BMI values are the same for both sexes and are considered to be age independent (24). However, the different body proportions and body composition of different populations are responsible for discrepancies between BMI and the degree of fatness (24). Increases in BMI are associated with a continuous increase in health risks, although the interpretation of BMI-levels in relation to risk may differ for different populations (25-27).

Due to the growing evidence that the associations between BMI and body composition differ across populations, especially in Asian and Pacific populations, the health risks may increase below the cutoff point of 25 kg/m<sup>2</sup> that defines overweight in the current WHO classification, has already triggered an intense debate (24). These debates have led to the convention of the WHO Expert Consultation on BMI in Asian populations (Singapore, 8-11 July, 2002) (28).

They decided that the proportion of Asian people with a high risk of type 2 diabetes and cardiovascular disease is substantial at BMI's lower than the existing WHO cutoff point for overweight (= 25 kg/m<sup>2</sup>). The variety of the cutoff point for observed risk ranges from 22 kg/m<sup>2</sup> to 25 kg/m<sup>2</sup> in different Asian populations and for high risk, it varies from 26 kg/m<sup>2</sup> to 31 kg/m<sup>2</sup>. The WHO Expert Consultation recommended that the current WHO BMI cutoff points (see table 1) should be retained as the international classification (28).

The cutoff points 23, 27.5, 32.5 and 37.5 kg/m<sup>2</sup> were added as additional cutoffs for public health actions (24). The Consultation recommended that countries should use all actual categories (i.e. 18.5, 23, 25, 27.5, 30, 32.5 kg/m<sup>2</sup>, and in many populations, 35, 37.5, and 40 kg/m<sup>2</sup>) for reporting purposes, to permit international comparisons (24).

**Table 1 BMI cutoff points and its risk to comorbidities defined by the WHO [adapted from (4)]**

Classification	BMI [kg/m <sup>2</sup> ] cutoff points	Additional cutoff points	Risk of comorbidities
Underweight	< 18.50		Low
Severe thinness	< 16.00		
Moderate thinness	16.00 - 16.99		
Mild thinness	17.00 - 18.49		
Normal range	18.50 to 24.99	18.50 - 22.99 23.00 - 24.99	Average
Overweight	≥ 25		
Pre-obese	25.00 to 29.99	25.00 - 27.49 27.50 - 29.99	Increased
Obese	≥30		
Obese class I	30.00 to 34.99	30.00 - 32.49 32.50 - 34.99	Moderate
Obese class II	35.00 to 39.99	35.00 - 37.49 37.50 - 39.99	Severe
Obese class III	≥ 40.0		Very severe

Aside the calculation of the BMI, there are some other measurements commonly used in lager studies including waist circumference, hip circumference, waist-hip ratio (WHR), sagittal abdomen diameter, bioelectrical impedance analysis, skinfolds and dual-energy X-ray absorptiometry. Those measurements were briefly discussed in the chapter 1.9.

### **1.5 BMI and disease**

The BMI indicates a somewhat stronger yet still moderate association with body fat and disease risk compared to estimates based on stature and body mass (29, 30). Increases in BMI are associated with increased risk of hypertension, dyslipidemia, insulin resistance, type 2 diabetes, coronary artery disease, deep venous thrombosis, stroke, gallbladder disease, haemorrhoids, hernia, osteoarthritis and musculoskeletal disorders, sleep apnoea and respiratory problems, Alzheimer's disease, renal disease, furthermore cancers of the endometrium, cervical, breast, prostate and colon (31-34).

## **1.6 BMI and mortality**

Large population surveys have shown that BMI is positively related to morbidity and mortality (35, 36).

In a recently published review evidence from 97 studies, with a total of 2.88 million people and 270.000 deaths, assessed the relationship between BMI standard categories [normal weight (BMI of 18.50 to 24.99), overweight (BMI of 25.00 to 29.99), obesity (BMI of  $\geq 30$ ), grade 1 obesity (BMI of 30.00 to 34.99), and grades 2 and 3 obesity (BMI of  $\leq 35$ )] and overall mortality (37).

Compared to normal weight, obesity of all grades and obesity grades 2 and 3 were associated with higher all-cause mortality [HR for all grades combined: 1.18 (95% CI 1.12-1.25), for grade 2 and 3 obesity 1.29 (95% CI, 1.18-1.41)]. Grade 1 obesity was not associated with a significant higher mortality [HR 0.95 (95% CI, 0.88-1.01)]. Moreover overweight was associated with significant lower all-cause mortality [HR 0.94 (95% CI, 0.91-0.96)] (37).

## **1.7 BMI and body fat**

However, despite the fact that it is mostly the excess adipose tissue (not excess body weight) that is associated with increased health problems (38) very limited data exist regarding its association between body fat and total and cardiovascular mortality (39).

Although BMI is correlated ( $r = 0.60-0.82$ ) with percentage total body fat (TBF%) (40), there is a lack of research regarding the usefulness of BMI as a surrogate for TBF%, especially in young adults and athletes.

The BMI classification system is derived from cut points obtained from the general population and may not be specific to subgroups such as physically active

individuals (e.g. athletes) and young physically inactive individuals (9). In general, there is little consensus on the use of body fat percentage criteria to define obesity or excess body fat levels (41).

The National Institute of Health (NIH), American Association of Clinical Endocrinology (AACE), and American College of Endocrinology (ACE) define obesity as an excess of total body fat, specifically body fat that is 25% of total body weight in men and 30-35% in women (42, 43).

The American College of Sports Medicine (ACSM) recommended on the basis of data reported by Gallagher and colleagues (44) a TBF% over 33% in women and 20% in men as acceptable cut points for overfatness in athletes (45). In developing these equations, the authors used race, age and sex as predictor variables to help explain the model. Recently published TBF% cutoffs from Heo et al. (46) which are comparable with those of Gallagher et al. tend to be higher, especially in younger groups regardless of age, sex and ethnicity. Heo et al. assume that 23-25% TBF in men and 35-37% TBF in women corresponds to a BMI of 25 kg/m<sup>2</sup> in young African Americans and white adults (aged 18-29) as shown in table 2.

**Table 2 Cutoffs of percentage of body fat in reference to BMI cutoffs (kg/m<sup>2</sup>) in women and men [data from (44, 46)]**

	<b>BMI cutoff</b>	<b>Gallagher</b> 20-39 [y]	<b>Heo</b> 18-29 [y]
<b>Women</b>	[kg/m <sup>2</sup> ]	[%]	
Underweight	<18.5	21	25-27
Overweight	≥25	33	35-37
Obesity	≥30	39	40-42
<b>Men</b>	[kg/m <sup>2</sup> ]	[%]	
Underweight	<18.5	8	12-15
Overweight	≥25	20	23-25
Obesity	≥30	25	28-29

## **1.8 Limitations of BMI**

The BMI classification system is derived from cut points obtained from the general adult population and may not be specific to subgroups such as physically active individuals (e.g. athletes) and young physically inactive individuals (9). Also under conditions such as infancy, ageing, racial differences and special clinical circumstances the BMI can provide misleading information about the body fat content due to the different proportion of lean body mass and hydration status in these subgroups (10).

The data of this study underline the fact that the BMI can not distinguish between athletes and non-athletes in males as well as in females (see figure 1).

The BMI does not discriminate between the different components of the body and cannot describe the regional fat distribution (2, 8, 47), as the lipometer does.

Figure 3 shows the comparison between a male athlete (age 19.9 y, height 1.72 m, weight 62 kg, BMI 21.0) and a male non-athlete (age 19.9 y, height 1.72 m, weight 59 kg, BMI 19.9) and figure 4 the comparison between a female athlete (age 17.1 y, height 1.72 m, weight 57 kg, BMI 19.3) and a female non-athlete (age 18.8 y, height 1.72 m, weight 59 kg, BMI 19.9 kg/m<sup>2</sup>).

BMI of the male athlete is higher than the BMI of the male non-athlete, nevertheless the athlete's subcutaneous adipose tissue (SAT) is lower at all body sites in comparison to the male non-athlete (see figure 3).

In case of the females the BMI is minimal lower (0.6 kg/m<sup>2</sup>) in the female athlete compared to the female non-athlete, however the female athlete's SAT is much lower compared to the female non-athlete (see figure 4).

Compared with the general adult population, the influence of large muscle mass on the BMI in athletes and young adults may misclassify these individuals as overweight and obese (9, 48).

In fact that the excess adipose tissue is the cause of the co-morbid conditions and not the excess weight (38), the use of TBF% and subcutaneous fat patterns may be more effective than BMI in assessing fatness and obesity in physically active individuals and young adults.

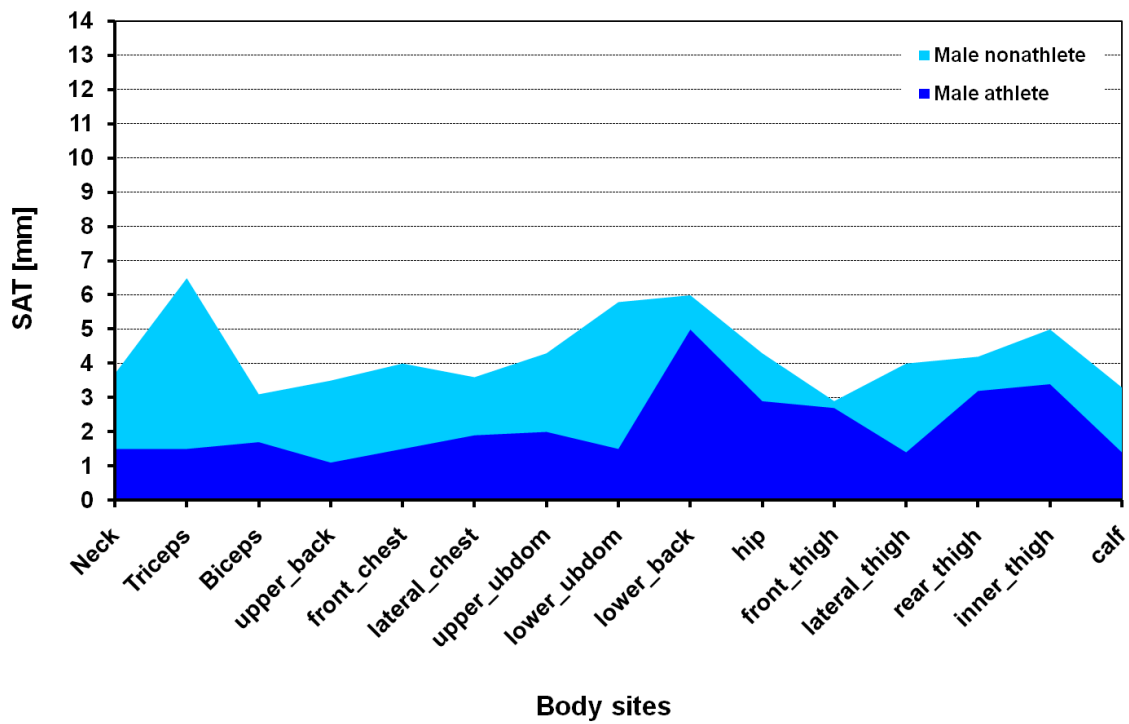


Figure 3 SAT-Top plot for a male athlete and his not physically active control, showing the SAT-differences of the 15 top-down sorted body sites in millimeters

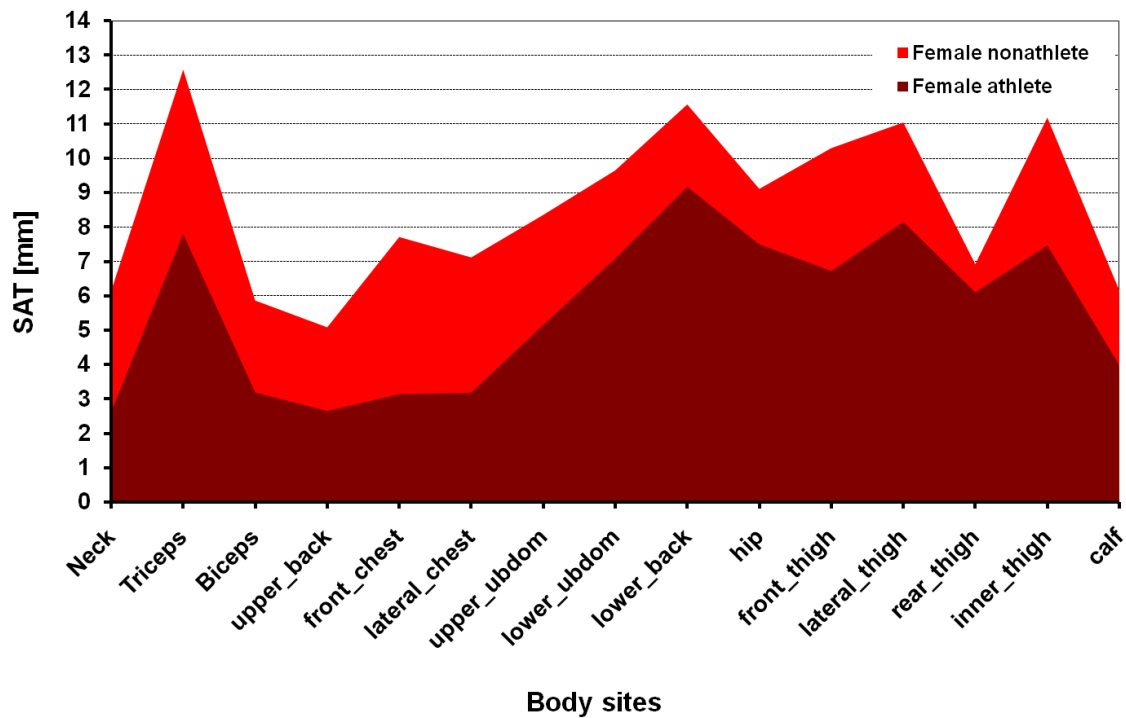


Figure 4 SAT-Top plot for a female athlete and her not physically active control, showing the SAT-differences of the 15 top-down sorted body sites in millimeters

## **1.9 Lipometer**

The computerized optical device named the Lipometer (Moeller Messtechnik, Graz, EU patent number 0516251) allows a non-invasive, quick, precise and safe determination of the thickness of subcutaneous adipose tissue (SAT) layers at any chosen site of the human body. The technical characteristics of the measurement system and a validation of the results using computerised tomography as the reference method have already been published (49).

Fifteen anatomically well-defined body sites (see table 3 and figure 5) have been specified to standardise Lipometer measurements (50), providing a subcutaneous adipose tissue topography (SAT-Top) of the subject. In adults the reliability of the SAT-Top method produced coefficients of variation ranging from 1.9% (front chest) to 12.2% (rear thigh) (51). SAT-Top includes the complete subcutaneous fat distribution information of a subject. Previous results have confirmed the importance of SAT-Top measurements in the fields of obesity, nutrition and metabolic disorders in children (52) and adults (53-56).

**Table 3 Description of the 15 specific body sites and their coefficient of variation [adapted from (57)]**

<b>Body site</b>	<b>Description</b>	<b>Coefficient of variation (n = 10) [%]</b>
1-Neck	40mm on the right side of the spine at vert. 7 (prominens)	4.8
2-Triceps	in the middle of the line between acromion and olecranon	6.0
3-Biceps	at the maximum girth of biceps muscle at front centreline of the arm	9.5
4-Upper back	below the scapula angulus inferior	2.2
5-Front chest	in the middle of the line between acromion and xiphoid process of sternum	1.9
6-Lateral chest	in the midaxillary line at the 10th rib	3.2
7-Upper abdomen	from the middle of the line between the xiphoid process of sternum and umbiculus 40 mm on the right side	3.8
8-Lower abdomen	40 mm on the right side of the umbiculus	3.2
9-Lower back	on the right side of the spine above the line of the iliac crest	6.7
10-Hip	in the midaxillary line above the iliac crest	3.7
11-Front thigh	in the middle of the front centreline between inguinal crease and patella	5.5
12-Lateral thigh	at the same height as 11, at lateral centreline of the leg	5.6
13-Rear thigh	at the same height as 11, at rear centreline of the leg	12.2
14-Inner thigh	at the same height as 11, at inner centreline of the leg	4.2
15-Calf	at the maximum girth of calf muscle (gastrocnemius) at rear centreline of the leg	5.8

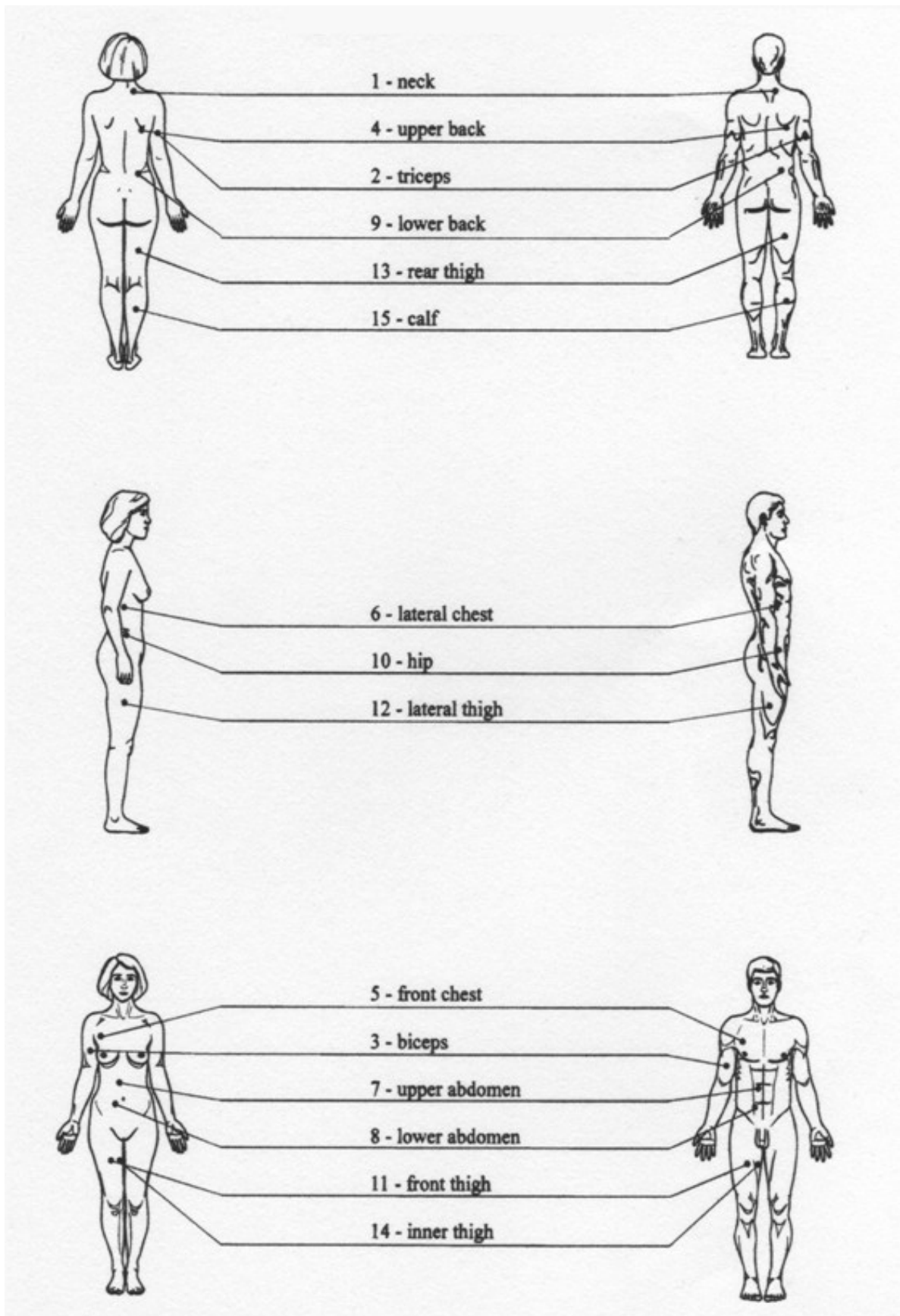


Figure 5 The 15 body sites used for the SAT-Top [adapted from (50)]

## 2 Objectives

Irrespective of the known limitations of BMI, it is still used to assess fatness in young adults (58) and athletes (59). But the accuracy of using the BMI as an indicator of body fat in these populations is questionable. As far as we know there has been no study that has assessed the relationship between BMI and SAT-Top in young athletes and non-athletes.

Therefore the purpose of this study was to prove our hypothesis that compared to BMI levels the subcutaneous fat patterns are a better screening tool to characterize fatness in athletes compared to non-athletes. A secondary aim of this study was to provide appropriate subcutaneous adipose tissue measuring points and cutoffs that allow a quick and precise way to discriminate between athletes and non-athletic controls.

### 3 Material and Methods

#### 3.1 Subjects

In this cross-sectional study the age, height, weight, BMI and SAT-Top were determined in 64 men (32 athletes and 32 non-active controls matched in age, height, weight and BMI) and 42 women (21 athletes and 21 non-active controls with comparable age and height). The female athletes had a significantly higher weight and BMI compared to the control females. Descriptive characteristics of the groups are presented in table 4 and 5.



Figure 6 Physique of a swimmer (left side) and a triathlete (right side) [adapted from (60, 61)]

Participants were subsequently separated into 2 groups:

- ⇒ **Athletes:** Twenty-three Swimmers (8 females and 15 males) and 30 triathletes (13 females and 17 males) were recruited from triathlon and swimming clubs in Graz (Austria) and Christchurch (New Zealand). They were between the ages of 15 and 30 years with at least 3 years training experience. The training and competition frequency was at least 2 hr/day, 6 days/week.

In a pre-test we investigated differences in body composition between swimmers and triathletes. We found no significant differences between the two groups, with the exception of the rear thigh measurement in women. Therefore we merged swimmers and triathletes to one group of athletes. Comparison of the swimmers and the triathletes physique see figure 6.

- ⇒ **Non-athletes:** Non-athletes were recruited via an advertisement. The subjects of the non-athletic group were aged between 15 and 30 years, non-smokers, were currently taking no medication and performing no more than one hour of exercise per week.

**Table 4 Descriptive statistics of the two male groups**

Personal parameters	Male nonathletes (n =32)		Male athletes (n = 32)		Significance of differences <sup>1</sup>
Age (y)	25.8 ± 5.6	(22.1- 27.7)	23.0 ± 13.2	(17.8-31.0)	n.s. <sup>2</sup>
Height (m)	1.80 ± 0.1	(1.75-1.82)	1.8 ± 0.1	(1.75-1.84)	n.s. <sup>3</sup>
Weight (kg)	72.3 ± 8.7	(66.3-75.0)	72.0 ± 8.5	(66.3-74.8)	n.s. <sup>3</sup>
BMI (kg/m <sup>2</sup> )	22.4 ± 1.4	(21.6-23.0)	21.8 ± 2.3	(20.7-23.0)	n.s. <sup>3</sup>
SAT-Top <sup>4</sup>					
Neck	3.7 ± 3.7	(2.5-6.2)	1.2 ± 0.6	(1.0-1.6)	p<0.001
Triceps	4.9 ± 3.0	(3.5-6.5)	2.1 ± 1.9	(1.5-3.4)	p<0.001
Biceps	3.0 ± 1.6	(2.1-3.7)	1.5 ± 0.6	(1.2-1.8)	p<0.001
Upper back	3.6 ± 2.3	(2.5-4.8)	1.5 ± 1.0	(1.1-2.1)	p<0.001
Front chest	3.8 ± 2.9	(2.8-5.7)	1.8 ± 1.2	(1.3-2.5)	p<0.001
Lateral chest	4.2 ± 3.2	(2.7-5.9)	1.7 ± 0.9	(1.1-2.0)	p<0.001
Upper abdomen	5.4 ± 4.5	(3.5-8.0)	2.1 ± 1.4	(1.6-3.0)	p<0.001
Lower abdomen	5.6 ± 5.5	(3.6-9.1)	2.5 ± 2.6	(1.4-4.0)	p<0.001
Lower back	6.4 ± 4.9	(3.8-8.7)	4.7 ± 3.6	(3.0-6.6)	p<0.01 <sup>3</sup>
Hip	6.3 ± 4.3	(4.5-8.8)	2.5 ± 3.7	(1.7-5.4)	p<0.001
Front thigh	3.2 ± 2.3	(2.5-4.8)	1.9 ± 1.0	(1.4-2.4)	p<0.001
Lateral thigh	4.0 ± 2.4	(3.1-5.5)	1.7 ± 1.2	(1.3-2.5)	p<0.001
Rear thigh	3.5 ± 2.4	(2.4-4.8)	1.7 ± 1.6	(1.4-3.0)	p<0.001
Inner thigh	4.9 ± 4.0	(3.8-7.8)	2.8 ± 1.3	(2.1-3.4)	p<0.001
Calf	3.0 ± 1.7	(2.2-3.9)	1.6 ± 0.9	(1.3-2.2)	p<0.001
Compartments (mm)					
Arms <sup>5</sup>	7.5 ± 4.0	(5.6-9.6)	3.7 ± 2.0	(3.2-5.2)	p<0.001
Legs <sup>6</sup>	19.3 ± 8.0	(15.8-23.8)	10.1 ± 3.9	(8.5-12.4)	p<0.001
Abdomen <sup>7</sup>	24.9 ± 18.8	(17.1-35.9)	12.1 ± 8.7	(8.5-17.2)	p<0.001
Trunk <sup>8</sup>	14.6 ± 10.9	(11.5-22.4)	6.5 ± 2.2	(5.6-7.8)	p<0.001
Total SAT <sup>9</sup>	68.3 ± 36.6	(52.9-89.5)	33.8 ± 13.4	(26.5-39.9)	p<0.001
TBF%	15.4 ± 4.7	(12.8-17.5)	10.2 ± 2.9	(8.5-11.4)	p<0.001

Data is Median ± interquartile range (1<sup>st</sup> to the 3<sup>rd</sup> quartile)

<sup>1</sup> By Mann-Whitney U test

<sup>2</sup> Not significant (P >0.05)

<sup>3</sup> By t-test for independent samples

<sup>4</sup> SAT thickness of 15 body sites in mm

<sup>5</sup> Body sites biceps + triceps

<sup>6</sup> Body sites front thigh + lateral thigh + rear thigh + inner thigh + calf

<sup>7</sup> Body sites upper abdomen + lower abdomen + lower back + hip

<sup>8</sup> Body sites neck + upper back + lateral chest + front chest

<sup>9</sup> Body sites 1-15

**Table 5 Descriptive statistics of the two female groups**

Personal parameters	Female nonathletes (n =21)		Female athletes (n = 21)		Significance of differences <sup>1</sup>
Age (y)	24.8 ± 2.6	(23.6-26.2)	21.7 ± 16.1	(17.1-33.2)	n.s. <sup>2</sup>
Height (m)	1.66 ± 0.1	(1.62-1.71)	1.7 ± 0.1	(1.64-1.73)	n.s. <sup>3</sup>
Weight (kg)	54.0 ± 6.8	(52.0-58.8)	60.0 ± 8.0	(55.0-63.0)	p<0.05
BMI (kg/m <sup>2</sup> )	19.9 ± 1.0	(19.7-20.7)	20.8 ± 2.1	(20.0-22.1)	p<0.05
SAT-Top <sup>4</sup>					
Neck	5.8 ± 3.6	(4.0-7.6)	2.4 ± 2.1	(1.5-3.6)	p<0.001
Triceps	12.5 ± 4.4	(9.9-14.3)	7.9 ± 2.0	(7.0-9.0)	p<0.001
Biceps	5.3 ± 3.6	(4.0-7.6)	3.2 ± 2.0	(2.2-4.2)	p<0.001 <sup>3</sup>
Upper back	4.9 ± 2.6	(3.7-6.3)	2.3 ± 1.4	(1.7-3.1)	p<0.001
Front chest	8.6 ± 5.9	(4.4-10.3)	2.7 ± 2.6	(1.8-4.4)	p<0.001
Lateral chest	6.3 ± 5.4	(4.6-10.0)	2.2 ± 3.3	(1.4-4.7)	p<0.001
Upper abdomen	7.4 ± 5.2	(5.5-10.7)	3.8 ± 5.1	(2.6-7.7)	p<0.01
Lower abdomen	10.2 ± 6.9	(6.0-12.9)	6.3 ± 4.7	(4.2-8.9)	n.s. <sup>3</sup>
Lower back	11.4 ± 5.2	(8.6-13.8)	9.1 ± 3.8	(7.2-11.0)	p<0.05 <sup>3</sup>
Hip	8.5 ± 6.6	(5.4-12.0)	7.1 ± 7.7	(3.4-11.1)	n.s. <sup>3</sup>
Front thigh	10.3 ± 3.5	(8.0-11.5)	6.9 ± 3.6	(4.6-8.2)	p<0.001
Lateral thigh	10.4 ± 2.8	(9.9-12.7)	8.0 ± 3.4	(6.5-9.9)	p<0.01 <sup>3</sup>
Rear thigh	7.2 ± 1.9	(6.1-8.0)	5.8 ± 2.7	(5.0-7.7)	n.s. <sup>3</sup>
Inner thigh	11.2 ± 2.8	(9.8-12.6)	7.4 ± 5.0	(5.4-10.4)	p<0.001 <sup>3</sup>
Calf	6.3 ± 2.3	(4.8-7.1)	3.5 ± 2.8	(2.5-5.3)	p<0.001 <sup>3</sup>
Compartments (mm)					
Arms <sup>5</sup>	17.4 ± 5.2	(15.7-20.9)	11.1 ± 2.3	(10.1-12.4)	p<0.001 <sup>3</sup>
Legs <sup>6</sup>	46.6 ± 9.1	(40.2-49.3)	30.9 ± 16.1	(24.2-40.3)	p<0.001 <sup>3</sup>
Abdomen <sup>7</sup>	40.5 ± 21.4	(25.7-47.1)	26.5 ± 23.2	(16.4-39.6)	p<0.05 <sup>3</sup>
Trunk <sup>8</sup>	25.6 ± 17.1	(17.1-34.2)	10.2 ± 6.4	(7.8-14.2)	p<0.001
Total SAT <sup>9</sup>	133.7 ± 48.7	(102.1-150.7)	78.8 ± 42.1	(62.8-104.9)	p<0.001 <sup>3</sup>
TBF%	30.2 ± 5.5	(27.2-32.7)	26.9 ± 4.7	(24.8-29.5)	p<0.01 <sup>3</sup>

Data is Median ± interquartile range (1<sup>st</sup> to the 3<sup>rd</sup> quartile)

<sup>1</sup> By Mann-Whitney U test

<sup>2</sup> Not significant (P > 0.05)

<sup>3</sup> By t-test for independent samples

<sup>4</sup> SAT thickness of 15 body sites in mm

<sup>5</sup> Body sites biceps + triceps

<sup>6</sup> Body sites front thigh + lateral thigh + rear thigh + inner thigh + calf

<sup>7</sup> Body sites upper abdomen + lower abdomen + lower back + hip

<sup>8</sup> Body sites neck + upper back + lateral chest + front chest

<sup>9</sup> Body sites 1-15

## **3.2 Ethics statements**

The participants provided their written informed consent to the study after receiving a thorough explanation of the study and its requirements. For participants under the age of 18, two informed consents were provided, one for their caretaker and one for themselves, as required by the local ethics committee. The procedures used in this study were in accordance with the Declaration of Helsinki and were approved by the ethics committee of the medical university of Graz (IRB00002556) (EC-number 19-054 ex 07/08).

## **3.3 Anthropometric Measurements**

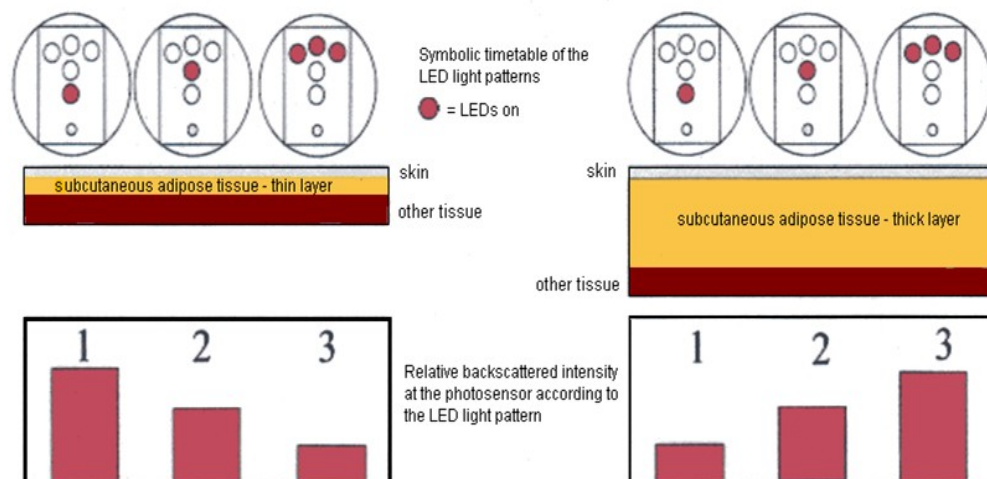
### **3.3.1 Measurement of weight, height and calculation of BMI**

Subjects wore light clothing (e.g. shorts and a light top) and no shoes during the measurements. Standing height was measured to the nearest 0.1 cm using a portable calibrated stadiometer (SECA®-220, Hamburg, Germany). Body mass was measured to the nearest 0.01kg using calibrated electronic scales (Soehnle® 7700, Murrhardt, Germany), BMI was calculated as body mass (kg) divided by height (m) squared.

### **3.3.2 Measurement of SAT-Top**

The optical Lipometer device was applied to measure the thickness of SAT in millimetres at 15 well-defined body sites distributed from neck to calf (see table 3 and figure 5). Measurements were performed on the right side of the body while subjects were in an upright standing position by a qualified technician. This set of measurement points defines the SAT-Top of each subject (50). The complete SAT-Top measurement cycle for one subject lasts about two minutes. The sensor head of the optical Lipometer device consists of a set of light emitting diodes ( $\lambda =$

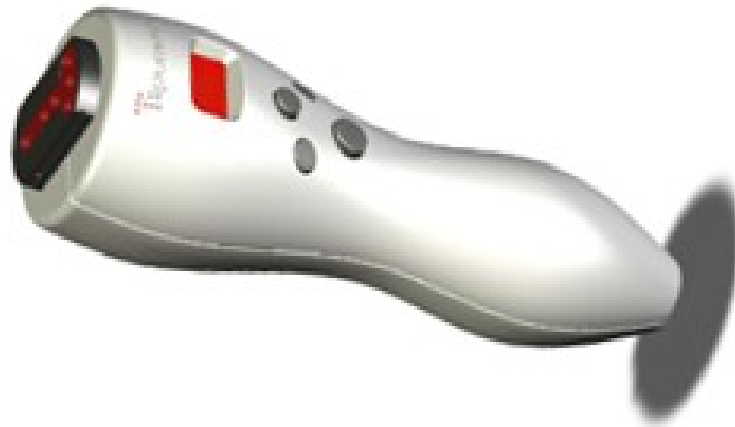
660 nm; light intensity, 3000 mcd) as light sources and a photodetector. During measurement, the sensor head is held perpendicular to the selected body site. The diodes illuminate the SAT-layer and the photodetector measures the corresponding light intensities back-scattered (see figure 7) (57).



**Figure 7 Ratio of backscattered intensities according to LED position switched on (1,2,3) in a thin SAT layer (left side) and thick layer (right side). [adapted from (57)]**

The resulting light pattern values of a measured body site were calculated to absolute SAT layer thickness (in mm) using Computer tomography (CT) as the reference method. The level of agreement between CT and the Lipometer has been found to be very high (correlation coefficient of  $r=0.99$ , with a regression line  $y=0.97x+0.37$ , and no systematic deviation of the Lipometer measurements from the CT results [Bias= $0.0\pm 1.42$ ]) and small limits of agreement [ $-2.78, +2.78$ ]. The use of this algorithm allows the conversion of the Lipometer data in mm thickness of any measured SAT thickness from 0 to 50 mm.

Due to the rectangular surface of the sensor head (38 x 25 mm) (see figure 8) the pressure on the particular body site during the measurement can be neglected. (51, 57)



**Figure 8 Lipometers sensor head [adapted from (62)]**

The current results of all 15 measured body sites of a person are presented on the screen in comparison to age- and sex specific reference values, based on the data of 30.000 men, women and children (age from 7 to 80) (see figure 18 in the appendix).

According to the fat distribution we can distinguish between a pear-profil (gynoid fat pattern) and an apple-profil (android fat pattern).

To calculate the subcutaneous fat mass (FM) and the lean body mass (LBM) a validated algorithm by Total Body Electrical Conductivity (TOBEC) (57) and Dual Energy X-Ray Absorptiometry (DEXA) (63) is used. The algorithm is also used to determine the absolute (in kg) and the relative (%) content of total body fat (TBF).

### ***3.4 Record of training extent***

To record the extent of training and competition load in individuals, structured questionnaires were used from which training volume in kilometres and hours per week was calculated. Dietary intake was obtained by using 24h-dietary recalls.

### 3.5 Statistics

Statistical calculations were performed by SPSS for Windows (version 17.0). Due to the distribution of the data the median, 1<sup>st</sup>Quartile (Q1), 3<sup>rd</sup>Quartile (Q3) and interquartile range (IQR = Q3 – Q1) were used for the descriptive analysis of the various variables. The normal distribution of the variables was tested using the Shapiro-Wilk test and the Kolmogorov-Smirnov test. Differences in the distributions of variables between athletes and non-athlete controls was tested by a Student's t-test for 2 independent samples (in case of normally distributed variables) and by a Mann-Whitney U-test for 2 independent samples (if variables were not normally distributed).

The 15 individual SAT-Top body sites listed in table 3 have been described previously (57) and can be summed to estimate regional fat mass.

- ⇒ **Arms** = biceps + triceps
- ⇒ **Trunk** = neck + upper back + lateral chest + front chest
- ⇒ **Abdomen** = upper abdomen + lower abdomen + lower back + hip
- ⇒ **Legs** = front thigh + lateral thigh + rear thigh + inner thigh + calf

To give information about the total amount of subcutaneous fat in these two groups, all 15 SAT layer thicknesses were summed (Total SAT). Furthermore, TBF% was calculated by equations, developed in a former study (64), using dual-energy X-ray absorptiometry (DXA) as reference method (64). To estimate Lipometer TBF% stepwise multiple regression analysis was applied, using the calculated DXA TBF% as the dependent variable. Using the 15 Lipometer SAT thicknesses together with age, height, weight and BMI as independent variables provided the best estimations of Lipometer TBF% for both genders with strong correlations to DXA TBF% (R=0.99 for males and R=0.95 for females). The limits

of agreement were -2.48% to +2.48% for males and -4.28% to +4.28% for females. For both genders a bias of 0.00% was determined (64).

The selectivity of measurement points was detected by receiver operating characteristic (ROC) curve analysis, which is a useful method for organizing classifiers and visualizing their performance. Two different a priori hypotheses can be specified: that either smaller or larger parameter values are associated with stronger evidence of positivity (= group of athletes). The area under the ROC curve is calculated and the result is expressed as an Area Index (AI). The higher sensitivity and specificity, the more the ROC-Curve shifts into the upper left corner of the graph (high discriminating power) (see figure 9) and the AI moves towards 1.0, consequently the selectivity between the groups is strong. Generally the AI can reach from 0.0 to 1.0 (= strongest selectivity). If the curve is near the diagonal (= AI 0.5) the selectivity is weak. An AI <0.5 shows that the a priori hypothesis should be changed (see BMI in figure 10). In the ROC curve, the x coordinate represents the specificity and the y coordinate shows the sensitivity.

The highest sensitivity and specificity were obtained at the optimal cutoff point estimated by the Youden index (65). This optimal cutoff value provides the best discriminating power between the group of athletes and their controls, whereby smaller values are associated more strongly with the group of athletes.

## 4 Results

Male athletes and non-athletes were similar in terms of age, height, weight and BMI, however, male athletes showed a 50.5% lower Total SAT thickness ( $33.8 \pm 13.4$  mm) compared to male non-athletes ( $68.3 \pm 36.6$  mm,  $p < 0.001$ ). All SAT layer thicknesses at the 15 body sites from neck to calf were significantly lower in the male athletes compared to the male non-athletes (see table 4). This was also the case for the additional variables (the four compartment measurements and TBF%).

Even though the female athletes had significantly higher BMI ( $p = 0.016$ ) and weight ( $p = 0.011$ ), their Total SAT thickness was 41.1% lower ( $78.8 \pm 42.1$  mm) compared to their non-athlete counterparts ( $133.7 \pm 48.7$  mm,  $p < 0.001$ ). SAT at all measured body sites, for all body compartments and TBF% was significant lower in the female athletes compared to the non-athletes except for the lower abdomen, hip and rear thigh (= gynoid fat pattern) (see table 5).

ROC curves and the corresponding area indices were calculated for height, weight, BMI, TBF%, Total SAT, SAT-layer thicknesses at all 15 body sites and for the 4 compartments. Optimal cutoff values were analysed for body sites with a  $p$ -value  $\leq 0.05$ . Results are presented in table 6 and 7, and show the area indices for these variables for the two assumptions that either small or large values provide stronger evidence for positivity (= athletes).

The best discriminators between male and female athletes and non-athletes are presented as ROC curves and Box plots in figure 9 - 12. In men the neck measurement (see figure 9) (AI=0.952, sensitivity = 96.9%, specificity = 84.4%, optimal cutoff value 2.3 mm) and the trunk compartment (AI=0.960, sensitivity = 84.4%, specificity = 96.9, optimal cutoff value 15.5 mm) provided the strongest discrimination power (90.6% [= 58 of 64 of the subjects were correctly classified as athletes or controls]). The data showed no significant difference between the BMI

of athletes and non-athletes (see table 6).

In women the upper back measurement (see figure 10) (AI=0.888, sensitivity = 81.0%, specificity = 95.2%, optimal cutoff value 3.3 mm) and the arms compartment (AI=0.923, sensitivity = 100.0%, specificity = 76.2%, optimal cutoff value 15.9 mm) provided the strongest discrimination power (88.1% [= 37 of 42 correctly classified subjects]). Female athletes had significantly higher BMI, nevertheless the BMI AI was low (AI=0.717, discrimination power: 52.4% [22 of 42 correctly classified subjects]) (see table 7).

**Table 6 Area indices and optimal cutoff values obtained from ROC curve analysis for height, weight, BMI, 15 specified SAT-layers, 4 Compartments, Total SAT, and TBF% of 32 male athletes and 32 male non-athletes**

Personal parameters	Area index <sup>1</sup>		P	Optimal cutoff <sup>2</sup> [mm]	Sensitivity [%]	Specificity [%]	Correctly classified cases
	H <sub>0</sub> : Small	H <sub>0</sub> : large					
Height (m)		0.500	n.s.				
Weight (kg)	0.500	-	n.s.				
BMI (kg/m <sup>2</sup> )	0.500	-	n.s.				
TBF%	0.903	-	<0.001	11.5	78.1	93.8	85.9% (55 of 64)
Total SAT <sup>9</sup>	0.914	-	<0.001	51.7	93.8	78.1	85.9% (55 of 64)
SAT-Top <sup>4</sup>							
Neck	0.952	-	<0.001	2.3	96.9	84.4	90.6% (58 of 64)
Triceps	0.853	-	<0.001	3.3	75.0	87.5	81.3% (52 of 64)
Biceps	0.901	-	<0.001	2.1	87.5	81.3	84.4% (54 of 64)
Upper back	0.929	-	<0.001	3.0	100.0	65.6	82.8% (53 of 64)
Front chest	0.889	-	<0.001	2.4	75.0	90.6	82.8% (53 of 64)
Lateral chest	0.914	-	<0.001	2.7	90.6	81.3	85.9% (55 of 64)
Upper abdomen	0.882	-	<0.001	4.2	96.9	68.8	82.8% (53 of 64)
Lower abdomen	0.844	-	<0.001	5.2	93.8	59.4	76.6% (49 of 64)
Lower back	0.698	-	<0.01	7.5	87.5	46.9	67.2% (43 of 64)

Hip	0.809	-	<0.001	4.2	68.8	81.3	75.0% (48 of 64)
Front thigh	0.831	-	<0.001	2.5	78.1	84.4	81.3% (52 of 64)
Lateral thigh	0.925	-	<0.001	2.8	87.5	87.5	87.5% (56 of 64)
Rear thigh	0.815	-	<0.001	2.1	56.3	93.8	75.0% (48 of 64)
Inner thigh	0.865	-	<0.001	3.7	81.3	81.3	81.3% (52 of 64)
Calf	0.821	-	<0.001	2.2	71.9	78.1	75.0% (48 of 64)
Compartments							
Arms <sup>5</sup>	0.907	-	<0.001	5.4	87.5	81.3	84.4% (54 of 64)
Trunk <sup>6</sup>	0.960	-	<0.001	15.5	84.4	96.9	90.6% (58 of 64)
Abdomen <sup>7</sup>	0.836	-	<0.001	19.8	84.4	75.0	79.7% (51 of 64)
Legs <sup>8</sup>	0.910	-	<0.001	8.2	93.8	78.1	85.9% (55 of 64)

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<sup>1</sup> There are two possible hypotheses ( $H_0$ ): that small/large values provide stronger evidence for positivity

<sup>2</sup> Optimal cutoff value estimated by Youden-Index (Youden, 1950)

<sup>3</sup> Not significant ( $p > 0.05$ )

<sup>4</sup> SAT thickness of 15 body sites in mm

<sup>5</sup> Body sites biceps + triceps

<sup>6</sup> Body sites front thigh + lateral thigh + rear thigh + inner thigh + calf

<sup>7</sup> Body sites upper abdomen + lower abdomen + lower back + hip

<sup>8</sup> Body sites neck + upper back + lateral chest + front chest

<sup>9</sup> Body sites 1-15

**Table 7 Area indices and optimal cutoff values obtained from ROC curve analysis for height, weight, BMI, 15 specified SAT-layers, 4 Compartments, Total SAT and TBF% of 21 female athletes and 21 female non-athletes**

Personal parameters	Area index <sup>1</sup>			Optimal cutoff <sup>2</sup> [mm]	Sensitivity [%]	Specificity [%]	Correctly classified cases
	H <sub>0</sub> : Small	H <sub>0</sub> : large	P				
Height (m)		0.500	n.s. <sup>3</sup>				
Weight (kg)		0.728	<0.05	66.0	95.2	9.5	52.4% (22 of 42)
BMI (kg/m <sup>2</sup> )		0.717	<0.05	18.8	4.8	100.0	52.4% (22 of 42)
TBF%	0.757	-	<0.01	30.5	100.0	47.6	73.8% (31 of 42)
Total SAT <sup>9</sup>	0.866	-	<0.001	83.6	61.9	100.0	81.0% (34 of 42)
SAT-Top <sup>4</sup>							
Neck	0.901	-	<0.001	4.8	90.5	71.4	81.0% (34 of 42)
Triceps	0.908	-	<0.001	10.4	95.2	76.2	85.7% (36 of 42)
Biceps	0.853	-	<0.001	3.8	71.4	85.7	78.6% (33 of 42)
Upper back	0.888	-	<0.001	3.3	81.0	95.2	88.1% (37 of 42)
Front chest	0.881	-	<0.001	4.1	76.2	85.7	81.0% (34 of 42)
Lateral chest	0.866	-	<0.001	3.3	71.4	95.2	83.3% (35 of 42)
Upper abdomen	0.746	-	<0.01	4.7	57.1	85.7	71.4% (30 of 42)
Lower abdomen	0.500	-	n.s.				
Lower back	0.689	-	<0.05	11.6	85.7	47.6	66.7% (28 of 42)

Hip	0.500	-	n.s.				
Front thigh	0.859	-	<0.001	9.5	90.5	66.7	78.6% (33 of 42)
Lateral thigh	0.824	-	<0.001	9.1	71.4	95.2	83.3% (35 of 42)
Rear thigh	0.500	-	n.s.				
Inner thigh	0.842	-	<0.001	9.6	71.4	85.7	78.6% (33 of 42)
Calf	0.825	-	<0.001	5.6	85.7	66.7	76.2% (32 of 42)
Compartments							
Arms <sup>5</sup>	0.923	-	<0.001	15.9	100.0	76.2	88.1% (37 of 42)
Trunk <sup>6</sup>	0.909	-	<0.001	13.9	76.2	95.2	85.7% (36 of 42)
Abdomen <sup>7</sup>	0.707	-	<0.05	34.9	71.4	66.7	69.0% (29 of 42)
Legs <sup>8</sup>	0.854	-	<0.001	44.5	90.5	66.7	78.6% (33 of 42)

<sup>1</sup> There are two possible hypotheses ( $H_0$ ): that small/large values provide stronger evidence for positivity

<sup>2</sup> Optimal cutoff value estimated by Youden-Index (Youden. 1950)

<sup>3</sup> Not significant ( $p > 0.05$ )

<sup>4</sup> SAT thickness of 15 body sites in mm

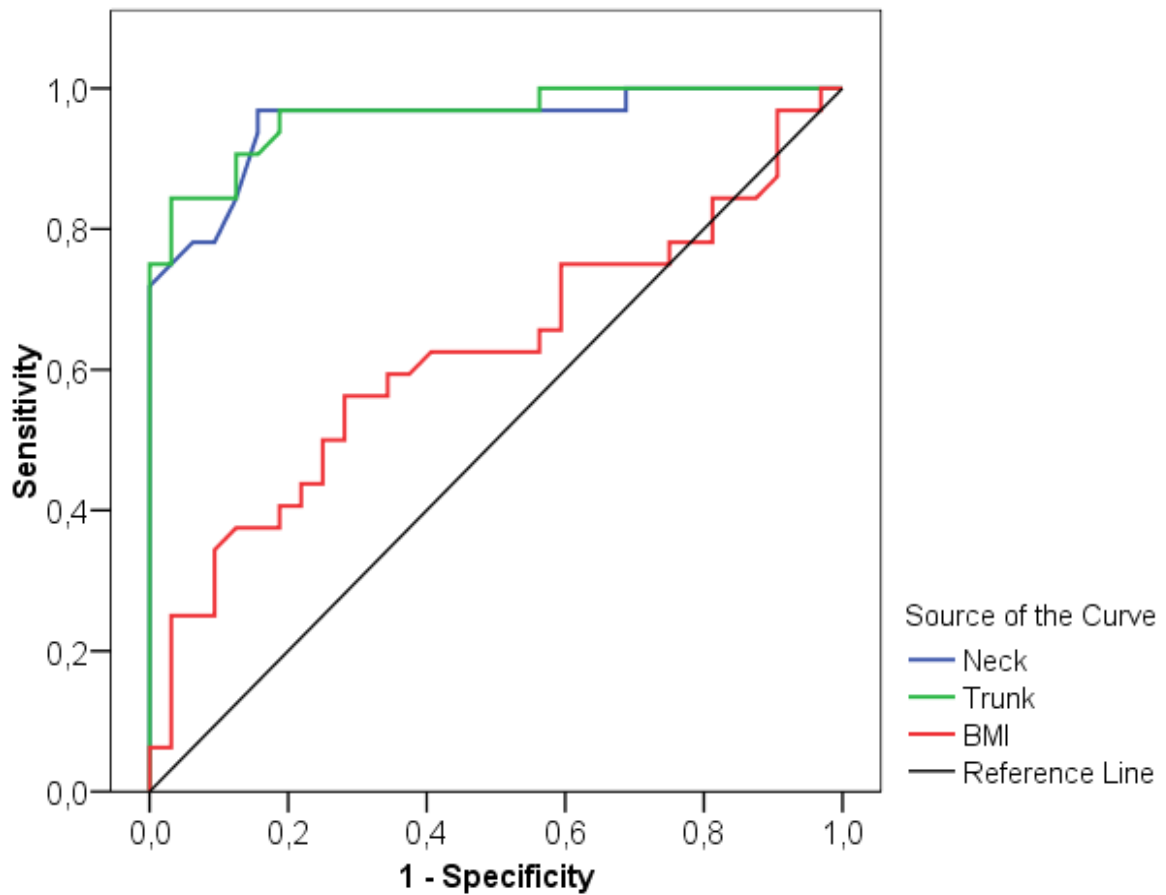
<sup>5</sup> Body sites biceps + triceps

<sup>6</sup> Body sites front thigh + lateral thigh + rear thigh + inner thigh + calf

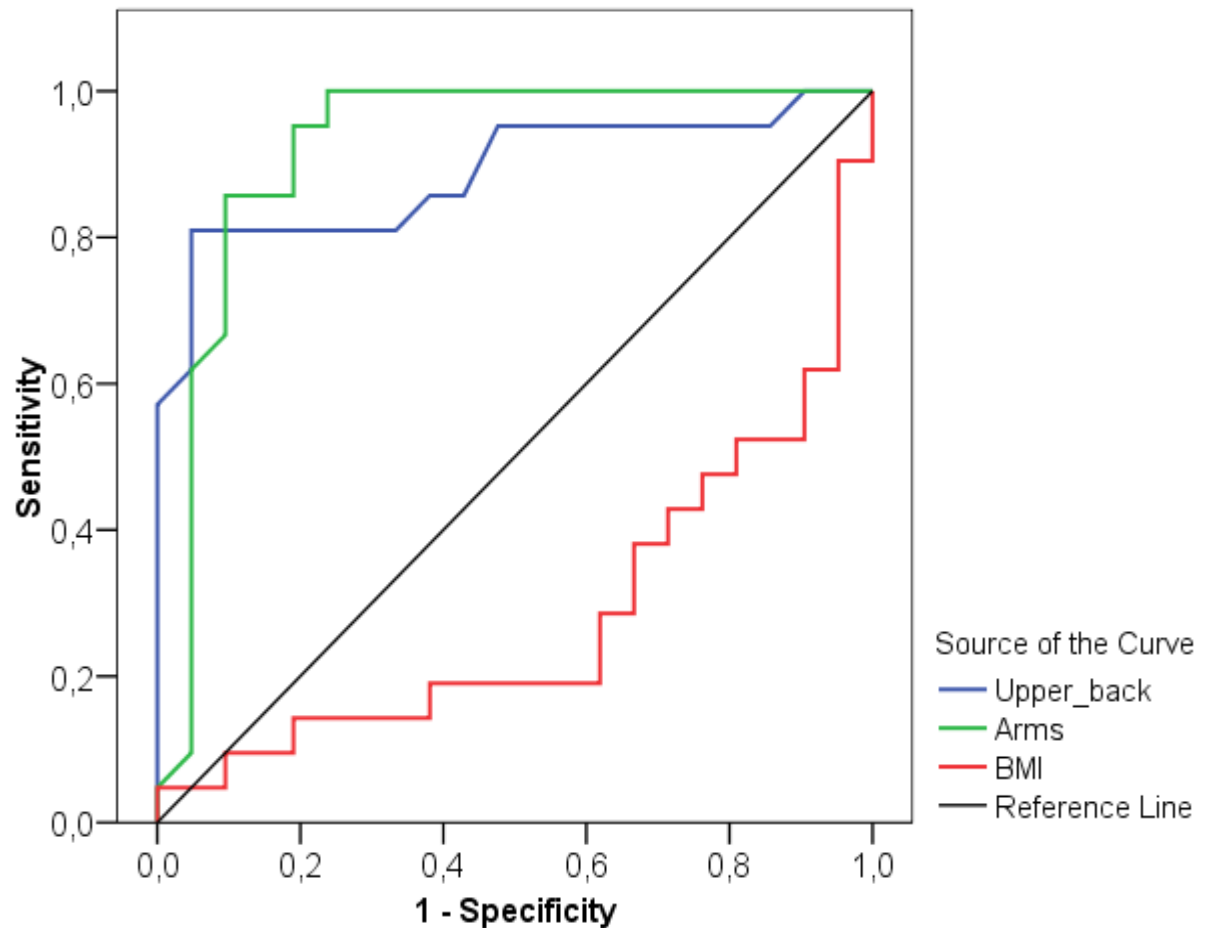
<sup>7</sup> Body sites upper abdomen + lower abdomen + lower back + hip

<sup>8</sup> Body sites neck + upper back + lateral chest + front chest

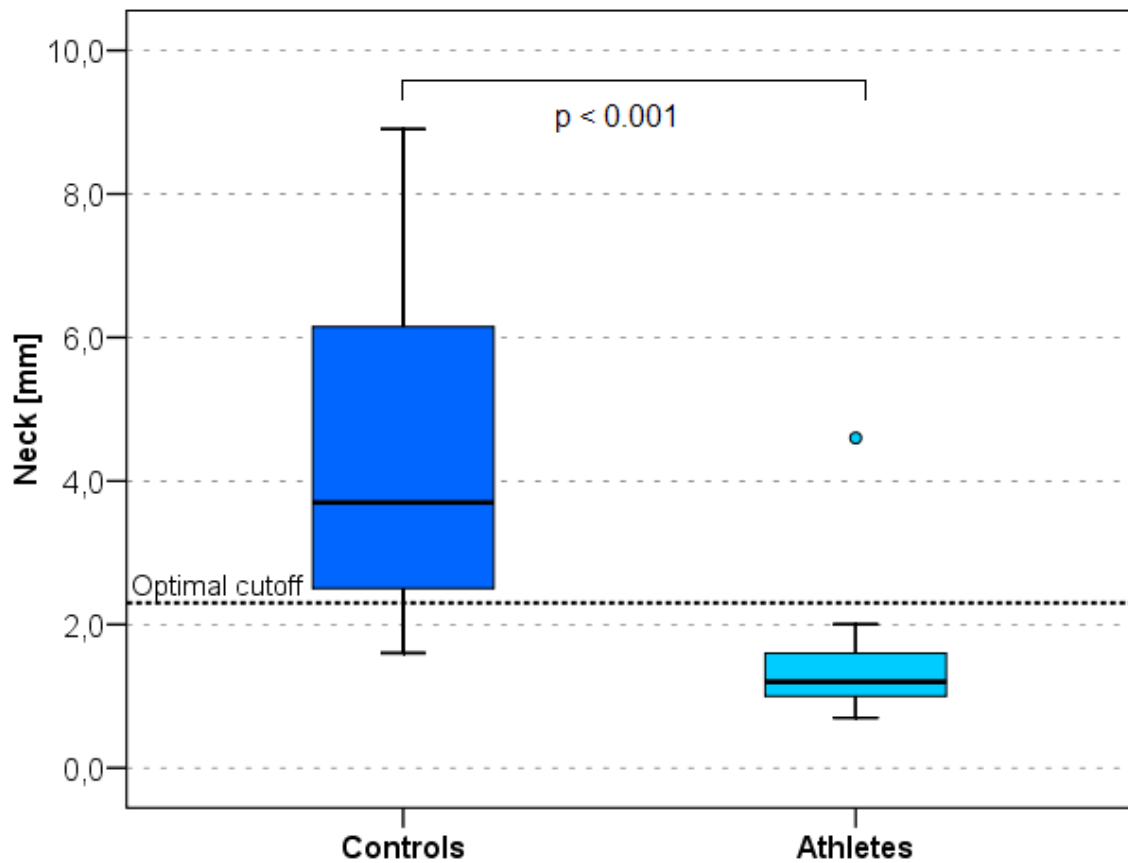
<sup>9</sup> Body sites 1-15



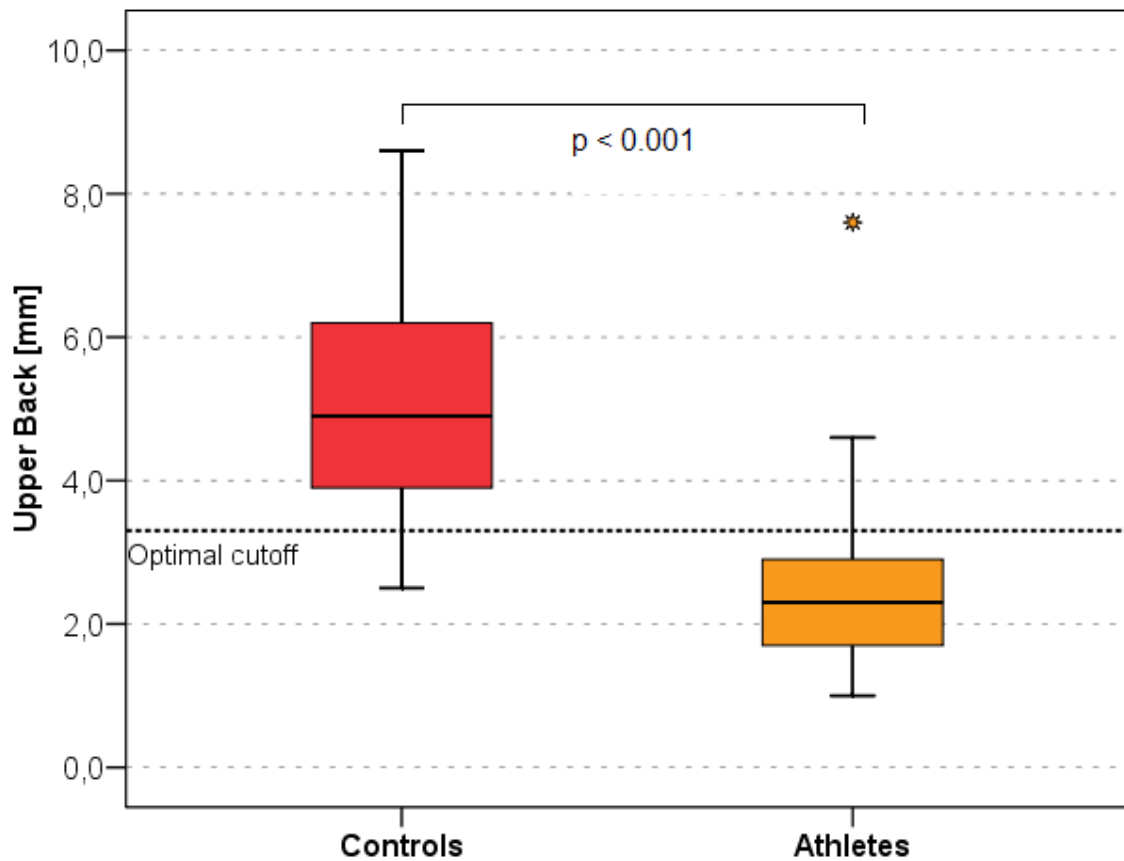
**Figure 9 Receiver-operator characteristics (ROC) curve for BMI, measuring point neck and compartment trunk of men.** The curve describes the association between sensitivity and specificity at different thresholds. ROC curves that approach the upper leftmost corner represent highly accurate classifiers.



**Figure 10 Receiver-operator characteristics (ROC) curve for BMI, measuring point upper back and compartment arms of women.** The curve describes the association between sensitivity and specificity at different thresholds. ROC curves that approach the upper leftmost corner represent highly accurate classifiers.



**Figure 11** Box plots of the measuring point neck of male controls and athletes which is the point with the highest discriminating power in men. The black horizontal lines represent the median, the box represents the 1<sup>st</sup> and 3<sup>rd</sup> quartile, the whiskers the 5<sup>th</sup> and 95<sup>th</sup> percentiles. Outliers are represented by dots. Optimal cutoff (= 2.3 mm) is marked by a dotted horizontal line.



**Figure 12** Box plots of the measuring point upper back of female controls and athletes which is the point with the highest discriminating power in women. The black horizontal lines represent the median, the box represents the 1<sup>st</sup> and 3<sup>rd</sup> quartile, the whiskers the 5<sup>th</sup> and 95<sup>th</sup> percentiles. Outliers are represented by dots. Optimal cutoff (= 3.3 mm) is marked by a dotted horizontal line.

#### **4.1 Other/Alternative Assessment Methods in Human Body Composition**

Measuring body composition in humans is usually in response to the need to describe either deficiencies or excesses of a component that is thought or known to be related to health risk (66). The currently available body composition measurements range from simple to complex and all of those methods having limitations and some degree of measurement error (66).

Despite of substantial progresses in the methods, there is still no gold standard for body fat assessment with accuracy better than 1% (67). Therefore it is important to select an appropriate method depending on the research question, study design and population under study (67). It is also very important to take in account, that strong association at the population level is not the same as a technique providing accurate, precise and reliable body composition data for an individual (67).

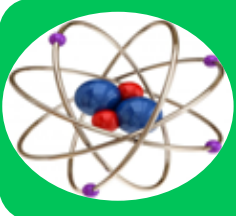
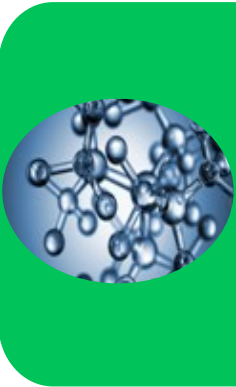
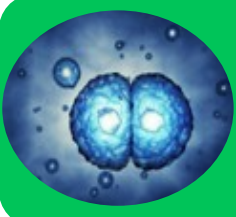
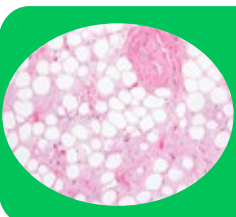

The most common methods for the assessment of body composition, with their advantages and limitations, will be mentioned in the next paragraphs.

Body mass can be seen as five distinct and separate but integrated levels (see table 8). The **first level** is the atomic level, which is based on the basic elements carbon, calcium, potassium, and hydrogen. **Second level**, the molecular one, is composed of the amounts of water, protein, and fat. Extracellular fluid and body cell mass presenting the **third level**, called the cellular level. The **fourth level**, also referred as tissue level, takes the amount of adipose, skeletal, and muscle tissues in account. At least the **fifth level** of body composition, the whole-body level, can be divided into regions such as appendages, trunk, and head. Those parts are usually described by anthropometric measures such as circumference, skinfolds, and length. [entire paragraph (13)]

The sum of all components at each of the five levels is equivalent for body mass. (3, 67)

The components of the different levels can not be measured directly. They rely on models of biological or physical relations, which are used to describe already known relation between a measured attribute and an unknown component, for example: total body water (TBW)/FFM =  $\sim 0.73$ ; total potassium/FFM = 60 mmol/kg. The relation between a certain indicator and a level component can also be established via a reference method and regression models (see chapter 1.9 reference methods). [entire paragraph (68)]

Table 8 Five Body Composition Levels [adapted from (67)]

	<p><b>1st Level: ATOMIC</b></p> <ul style="list-style-type: none"> <li>• <math>BM = H + O + N + C + Na + K + Cl + P + Ca + Mg + S</math></li> </ul>
	<p><b>2nd Level: MOLECULAR</b></p> <ul style="list-style-type: none"> <li>• <math>BM = FM + TBW + TBPro + Mo + Ms + CHO</math></li> <li>• <math>BM = FM + TBW + TBPro + M</math></li> <li>• <math>BM = FM + TBW + \text{nonfat solids}</math></li> <li>• <math>BM = FM + Mo + \text{residual}</math></li> <li>• <math>BM = FM + FFM</math></li> </ul>
	<p><b>3rd Level: CELLULAR</b></p> <ul style="list-style-type: none"> <li>• <math>BM = \text{cells} + ECF + ECS</math></li> <li>• <math>BM = FM + BCM + ECF + ECS</math></li> </ul>
	<p><b>4th Level: TISSUE-ORGAN</b></p> <ul style="list-style-type: none"> <li>• <math>BM = AT + SM + \text{bone} + \text{visceral organs} + \text{other tissues}</math></li> </ul>
	<p><b>5th Level: WHOLE-BODY</b></p> <ul style="list-style-type: none"> <li>• <math>BM = \text{head} + \text{trunk} + \text{appendages}</math></li> </ul>

AT = adipose tissue; BCM = body cell mass; BM = body mass; CHO = carbohydrates; ECF = extracellular fluid; ECS = extracellular solids; FFM = fat-free mass; FM = fat mass; M = mineral; Mo = bone mineral; Ms = soft tissue mineral; SM = skeletal muscle; TBPro = total body protein; TBW = total body water. Figure sources: picture for 1<sup>st</sup> and 3<sup>rd</sup> level adapted from: [www.wisegeek.com](http://www.wisegeek.com); picture for 2<sup>nd</sup> level: adapted from [www.medicine.ucsf.edu](http://www.medicine.ucsf.edu); picture for 4<sup>th</sup> level adapted from: [www.histologyolm.stevegallik.org](http://www.histologyolm.stevegallik.org); picture for 5<sup>th</sup> level adapted from: [www.potail.blogspot.com](http://www.potail.blogspot.com).

The molecular two-compartment model, including FM and FFM, is the most widely applied model in present body composition research (67).

Furthermore the techniques can be classified as being direct, like the cadaver dissection, or as being indirect, where a surrogate parameter is measured to estimate tissue or molecular composition. Moreover doubly indirect measures were used, those methods rely on an indirect measure that is used to predict another indirect measure (i.e. via regression equations). [entire paragraph (67)]

Considerable are also techniques like skinfolds, ultrasound, which sample the subcutaneous adipose tissue (SAT) at standardized sites and assume that there is some fixed and direct relation between this compartment and centralized body fat (67). Based on the standardized sites these methods provide an estimate of the total subcutaneous body fat (67). Also the Lipometer belongs to those methods.

#### **4.1.1 Reference Methods**

Reference methods are used to compare one method with another. Techniques, known as the most accurate techniques for body composition, were defined as standard and compared against the method that should be tested. Reference methods in body composition including cadaver dissection, multi-compartment model, MRI scanning, CT scanning, those methods are described in the following section. [entire paragraph (67)].

##### **4.1.1.1 Cadaver dissection**

The best data on human body composition come from cadaver studies. There are two different approaches behind this method. The chemical approach yields the amounts of water, protein, fat and mineral elements in the different tissues and the body as whole. The anatomical approach partitions the body into those components which are readily separated by dissection, including skin, muscle,

adipose tissue, bone and organs. In combination the two approaches yield data of the dissectable tissue. [entire paragraph (69)]

Although the cadaver dissection provides precious data, the high costs in time and for the cadavers, as well as considerable ethical barriers limiting its usability (69). The cadaver dissection is inapplicable for individual analysis, therefore practitioners have turned to other references, laboratory and field methods for estimating body composition (67).

#### Comment on the current study:

For the current study and in assessing body composition of athletes the cadaver dissection was/is no viable alternative.

#### **4.1.1.2 Multi-Component Models**

Multi-component models are the most commonly used body composition methods for the assessment of whole-body and segmental-body compartments (67). These models include fat mass, fat-free mass, total body water, body cell mass, extracellular water, intracellular water, bone mineral content and protein mass (70). They were deemed as best reference methods for estimation of body fat. Their precision and accuracy are within 1-2%. 6-, 5-, 4-, 3-compartment models are in use for the estimation of body fat, whereat the 4-compartment model, using body density, body water and bone mineral, is the most frequently used among them (67). The 4-compartment model is currently the leading reference method for body composition (67). 13 different 4-compartment equations were presented by Wang et al. (70), each of them with a different assumption for the various components. The form of those 13 equations is always equally composed (see Equation 2) (67).

$$\mathbf{FM = C_1 BV - C_2 TBW + C_3 Mo - C_4 BM}$$

**Equation 2 Basic 4-compartment equation [adapted from (67)]**

The 3-compartment model based on measures of body density and body water is able to estimate fatness within standard errors of estimate (SEEs) of 2.0-2.5% (67). Technical errors of estimating BV, TBW and Mo have been combined to yield a percentage of fat error of about 1% (67). Accuracy is in the order of 2% and even better when a 5-component model is used (67, 70).

Multi-compartment models are the most appropriate reference method to date, but nevertheless the complex analysis process, the use of expensive technologies and the lack of published normative data representing limitation of these methods (67).

#### Comment on the current study:

In the case of the current study the multi-compartment model was no feasible alternative because of the high time consume and the high costs and also the lack of published normative data for athletic populations. For further investigations it would be interesting to compare the estimate of total fat and SAT between the lipometer and a multi-compartment model.

#### **4.1.1.3 Imaging Methods**

Imaging methods are considered as the most accurate approaches for in vivo quantification of body composition at the tissue-organ level. Both Computer Tomography (CT) and Magnetic Resonance Imaging (MRI) produce high-resolution images. MRI and CT are the reference methods for calibration of field methods designed to measure adipose tissue (AT) and skeletal muscle in vivo. Those both are the only methods available for the estimation of internal tissues and organs in vivo. [entire paragraph (71)].

Compared to MRI CT has been shown to provide slightly more reliable and repeatable results and is more widely available than MRI (71). Nevertheless the major disadvantage of CT is the high radiation dose (see table 9) because its image acquisition is on the basis of X-rays, which are configured in a perpendicular plane to the supine individual under examination (67, 71).

#### 4.1.1.3.1 Computer Tomography (CT)

The basic CT system consists of an X-ray tube and a receiver that rotate in a perpendicular plane to the subject (see figure 13). The X-ray tube emits X-rays (0.1-0.2 Å, 60-120 kVp) that are attenuated as they pass through tissues, the remaining X-ray pattern is transmitted to a detector (e.g., film or a computer screen) for recording or further processing by a computer.

In contrast to conventional X-ray which produces only one picture of the body region under examination, CT produces many “slices” of internal organs and tissues. Out of the “slices” the CT produces absorption profiles of the object from many directions and reconstructs therefrom a volume structure. In contrast to normal X-rays the measured data do not consist out of two-dimensional pictures, but depicts a one-dimensional absorption profile. Based on computer assisted image reconstruction an image will be calculated. [entire paragraph (71)]

Conventional X-ray as well as CT uses the fact that different tissues have different radiographical density and differ therefore in their permeability of X-rays. The higher the density of the tissue, the more radiation will be attenuated and the lighter the depicted tissue appears (e.g. bones) (71).

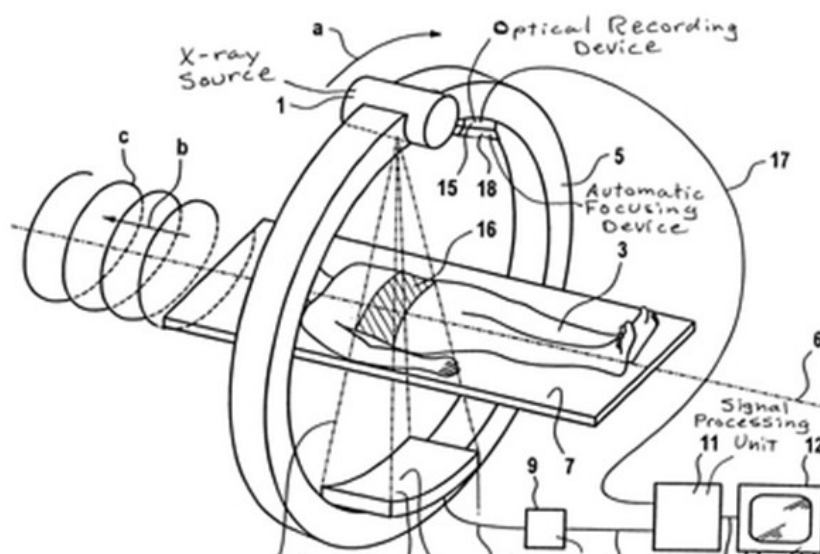


Figure 13 Computer Tomography Unit [adapted from (72)]

Conventional X-ray as well as CT uses the fact that different tissues have different radiographical density and differ therefore in their permeability of X-rays. The higher the density of the tissue, the more radiation will be attenuated and the lighter the depicted tissue appears (e.g. bones) (71). Skeletal muscle has a much higher density compared to adipose tissue that enables easy distinction and quantification of each (67).

Due to high radiation dose whole body scanning is infeasible, therefore most studies rely on interpolation between slices of measured composition. Overall CT is not a method for body composition assessment in daily routine. [entire paragraph (67)].

**Table 9 Radiation exposure [data from (66, 73-75)]**

	Effective dose [mSv]
Environmental radiation in Austria per year	0.6-1.8
conventional x-ray	0.02-1.5
DEXA whole body scan	0.0004-0.086
CT chest, abdomen and pelvis	9.9
CT chest	5.8
CT abdomen	5.3
CT abdomen and pelvis	7.1
CT head	1.5

#### **4.1.1.3.2 Magnetic Resonance Imaging (MRI)**

The procedure uses strong magnetic fields and radio waves to produce cross-sectional images of organs and internal structures in the body. The signal detected by an MRI machine varies depending on the water content (body tissue contains lots of water, and hence protons) and local magnetic properties of a particular area of the body, therefore different tissues or substances can be differentiated from each other. [entire paragraph (71)]

Instead of ionizing radiation the MRI is based on the interaction between hydrogen nuclei and the magnetic fields generated and controlled by the MRI system's instrumentation. Protons can be found in all biological tissues. Hydrogen protons have nonzero magnetic moments, which cause them to act like tiny magnets. In the MRI unit the field strength is mostly 15.000 times stronger than the earth's (e.g., 1.5 Tesla). If a subject is placed inside the magnet of an MRI unit consequently the protons align themselves with the magnetic field. [entire paragraph (71)]

Following the hydrogen protons are aligned in a known direction, a pulsed radio-frequency field is utilized to the body tissues causing a number of hydrogen protons to absorb energy. After switching off the radio frequency pulse, the protons gradually return to their original positions. During this process they release energy that is absorbed in the form of a radio frequency signal, which can be measured with receiver coils. This signal is used by the MRI system to create the cross-sectional images. The basis for high-quality MRI images are the differences created via the manipulation of the radio frequency parameters, which produce differences in relaxation times between different tissues and organs. [entire paragraph (67)]

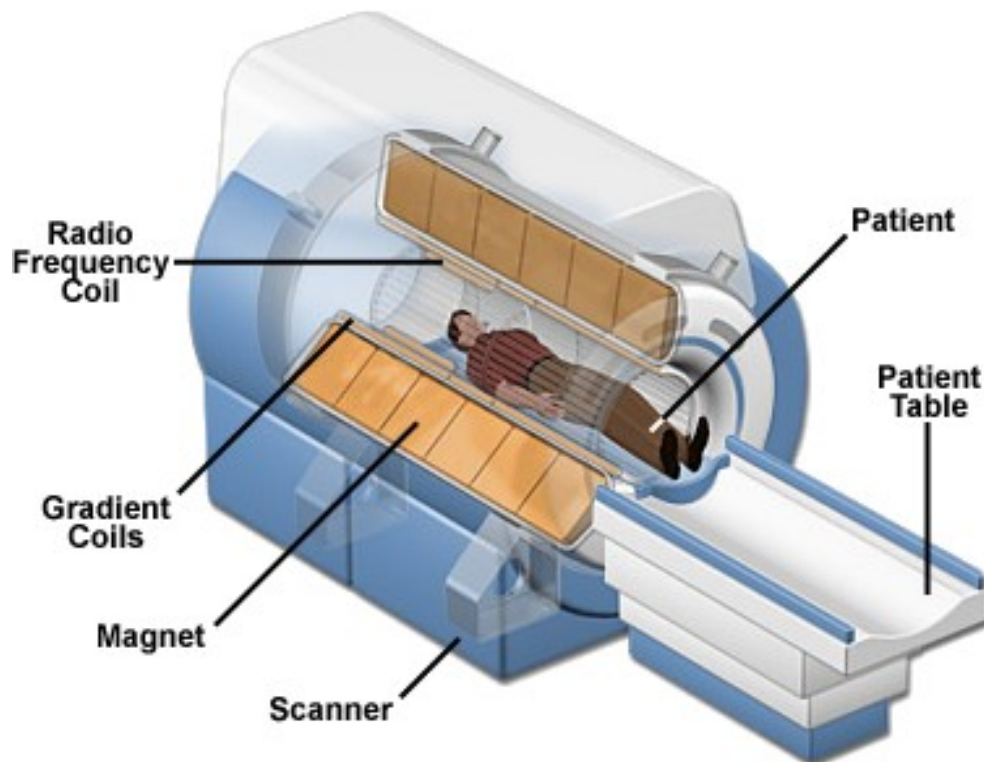


Figure 14 MRI Scanner Cutaway [adapted from (76)]

In most MRI devices, an electric current is passed through coiled wires to create a temporary magnetic field around a patient's body. In open-MRI devices, permanent magnets are used. Radio waves are sent from and received by a transmitter/receiver in the machine, and these signals are used to produce digital images of the area of interest (see figure 14) (75).

Limitations of MRI are high costs (acquisition costs and additional costs for data processing). Persons with claustrophobia cannot be scanned (67). Neither MRI nor CT is capable to measure very large persons ( $BMI > 40 \text{ kg/m}^2$ ), as the field-of-view for most MRI scanners is limited to  $48 \times 48 \text{ cm}$  (66). The MRI is limited in accuracy of measurement due to the pixel size of  $2 \text{ mm} \times 2 \text{ mm}$  in slices used in total-body scans (67).

#### Comment on the current study:

CT was no feasible method for this study because of high radiation dose of whole body scanning. Both methods, MRI and CT, are expensive, time consuming and not practicable for the daily routine, as the lipometer or anthropometry.

## 4.1.2 Laboratory Methods

Laboratory methods are body composition measurements used primarily for study purposes and seldom in the clinical field.

### 4.1.2.1 Dual-Energy X-Ray Absorptiometry (D(E)XA)

DXA is the most popular method for quantifying fat, lean, and bone tissues beneath the laboratory methods (3, 67).

DXA measurements were made with a total body scan that measured the attenuation of low-energy X-rays pulsed between 70 and 140 kV synchronously (77). The differential attenuation through the body allow the discrimination of total

body adipose and soft tissue, in addition to bone mineral content and bone mineral density. The person under examination has to lie on a scanning table, during the process maps the mass and composition of each pixel in terms of bone mineral, fat and fat-free soft tissue (67). A whole body scan lasts about 5 to 30 minutes (depending on the equipment) and exposes the subject to 0.04 – 0.86 mrem of radiation, which is equivalent to between 1 and 10% of a chest radiograph (3, 66, 67). Table 9 shows comparisons of radiation dose equivalent of different body composition methods.

DXA is a quick and convenient whole-body method that can be applied in humans of all ages (66). It is minimally influenced by water fluctuation (67) and it can be used to describe regional body composition and nutritional status in disease states and growth disorder (66). DXA is viewed as laboratory reference method and contributes to the bone mineral assessment for multi-component models (67).

Aside the small amount of radiation it should be taken into account that the method has some more limitations. The scanning bed has an upper limit and the whole-body field-of-view cannot accommodate individuals greater than ~192 cm

(67). For the most scanners the maximum weight is 120 kg (67). The tissue of very obese people can migrate beyond the available width of the scan area (66). Trunk thickness can influence the estimates of fat mass, increasing errors appear if individual's trunk thickness increases (66). Due to the radiation and moreover to the error of measurement, which limits the ability to detect small composition changes over time, a DXA scan should be done no more than four times per annum (67).

When DXA percent fat is compared against percent body fat using a multicomponent method, the SEE values typically range between 2.5% and 3.5% (77). Compared with CT and neutron activation analysis for assessing skeletal muscle a SEE of 1.6 kg and 4.4 kg was seen (77). The assessment of total and regional FFM is generally acceptable if total scanned mass equates to scale mass nevertheless it is not reliable in producing accurate fat estimates in lean athletes (67). Specific manufactures and models have been tested and found to have certain biases that may overestimate FFM (3).

Differences in hardware and software algorithms make it difficult to compare between different equipments. Future studies will clarify the limitations of its accuracy and lead to more standardized approaches in all populations (67, 77).

#### Comment on the current study:

A comparison between DXA, BMI and lipometer would be interesting. Although DXA is criticized for producing unreliable estimates of fat in lean populations, it is able to depict the zonal fat distribution as the lipometer does. Nevertheless it is no feasible field method as the lipometer, anthropometrie or BIA and DXA uses a little amount of radiation.

#### 4.1.2.2 Densitometry

At present there are two density measurements in use to estimate body composition from body density, underwater weighing (UWW) and air displacement plethysmography (ADP). Those two methods are based on a 2-compartment model which divides the body into FM and FFM and uses the inverse relationship between pressure and volume (Boyle's law) to derive body volume (l) for a subject (78). The density of the human body ( $D_b$ ), like any material, is equivalent to the ratio of its mass (MA) and volume (V) (79).

$$D_b = MA/V$$

Mass (MA) = body weight (kg)

Volume (V) = liters (l)

**Equation 3 Basic equation to calculate the density of an object**

Once the body mass, estimated from body weight, and volume are known, density can be calculated from equation 3 (79).

The Archimedes Principle is the basis of UWW which states that the buoyant force on a submerged object is equal to the weight of the fluid that the body displaces (80). Thus body volume (BV) is equal to the loss of weight in water, corrected for the density of water ( $D_w$ ) corresponding to the temperature of the water during the submersion (see equation 4) (79).

$$BV = (W_a - W_w)/D_w$$

( $W_a$  = weight in air;  $W_w$  = weight in water)

**Equation 4 to calculate the body volume**

Two extraneous volumes included in the total BV, air in the lungs and flatus in the gastrointestinal tract, have to be adjusted in the final calculation (79). Lipid is the only constituent of the body whose specific gravity is less than that of water (1.0) and its buoyant force is opposed by all other, denser constituents (bone, muscle) (67).

For UWW a tank of 3 m x 3 m x 3 m (4.0 m for subjects >2 m) and at least 2.3 m water depth is needed (79). The water in the tank should have a temperature of 32-35°C and the water quality has to be maintained through filtering and regular chemical treatments to maintain chlorine levels. Water pH must be maintained between 7.4 and 7.6 (79).

First the person weight is determined out of the water (80). Then the subject is seated on a submersible seat suspended from a load cell (79). The subject is encouraged to exhale maximally during submersion (79). This procedure is repeated three times and averaged (80).

UWW and ADP require estimation of residual lung volume (67). The residual volume is commonly measured using either the closed-circuit approach, which involves dilution and eventual equilibration of an inert tracer or indicator gas such as nitrogen oxygen, or helium, or the open-circuit approach, where nitrogen is “washed out” of the lungs during a specified period of oxygen breathing (79).

UWW determines body density, and percent body fat can be calculated using the Siri or Brozek formula, which are recently the simplest and most common fat-estimating formulas (79). Total error of estimating fat content is 0.0062 g/ml or equivalent of 2% fat (79).

UWW has been considered the gold standard for body composition assessment over a long time (79), nevertheless more sophisticated methods, such as DXA, may make underwater weighing obsolete in the future (80).

ADP follows a similar approach as UWW by measuring body volume in a sealed air capsule. ADP use air displacement technology (pressure-volume relationships) instead of water to estimate volume and density (79). Due to higher precision and accuracy compared with past techniques, the Bod Pod (Life Measurement Instruments, Inc., Concord, CA) is most frequently used for BV measurement since its introduction in 1995 (79).

This system consists of a measuring chamber that accommodates the subject and a reference chamber that contains instrumentation for measuring changes in pressure. The two chambers are linked via a airtight diaphragm which determines pressure changes between the chambers (see figure 15) (67, 81).

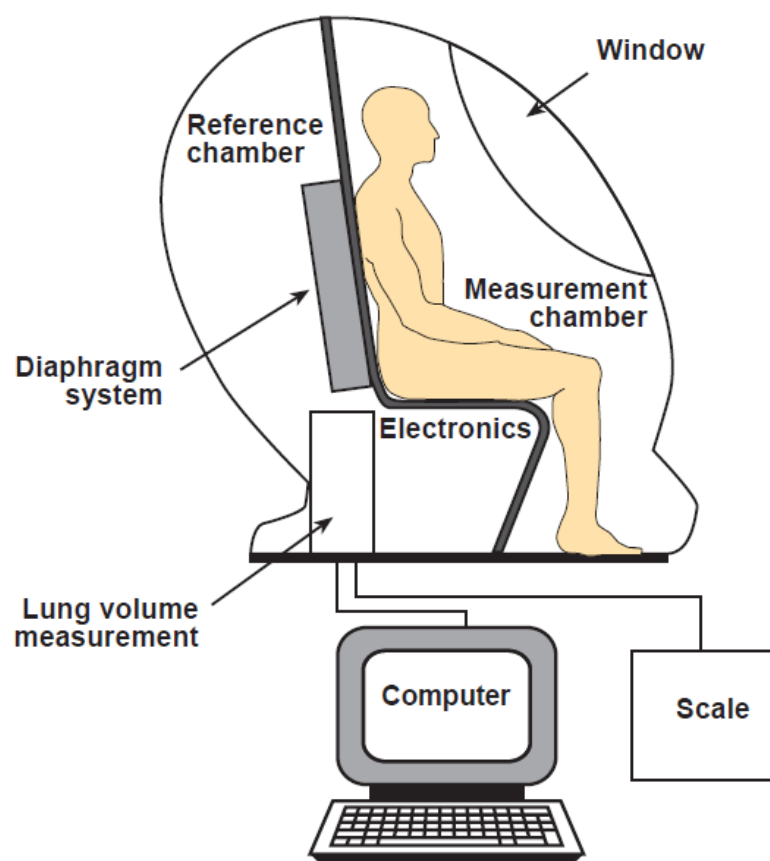


Figure 15 Composition of a Bod Pod [adapted from (78)]

The pressure ratio relationships between the chambers are inversely related and are characterized by Boyle's Law (see equation 5) (79, 81):

$$P_1/P_2 = V_2/V_1$$

**Equation 5 shows pressure ratio relationships in a Bod Pod chamber**

$V_1$  and  $P_1$  are the volume and pressure prior to subject entry into the test chamber and  $V_2$  and  $P_2$  are the volume and pressure while the subject is placed in the test chamber (81). Therefore, subject body volume will equal the volume of the test chamber before subject entry less the test chamber volume with the subject placed in the chamber (81). Percentage body fat is calculated using the Siri formula (78).

Before measurement the system is calibrated with a known volume of 49.550 l and the weighing scales are also calibrated against a known weight of 20 kg (67, 78). The subject is weight wearing minimal cloths, ideally swimwear and a cap, while it is placed for ~2 minutes in the fiberglass measuring chamber for volumetric measurement. The subject has to breathe normally during measurement and is then instructed to maximal in- and expiration for residual gas calculation. Due to the behavior of the air inside the chambers adjustments for thoracic gas volume and skin surface area are necessary. The behavior of air close to the skin surface is predicted by a surface area artifact, based on estimated body surface area. Such estimates may under- or overestimate an athlete's true surface area. Also the clothing can reduces test-retest reliability and leads to an underestimate of fatness. [entire paragraph (67)]

In humans reliability of the Bod Pod is good to excellent. In adults, the within-subject coefficients of variation (CVs) for percent body fat have ranged from 1.7% to 4.5% within a day. These CVs are within the range of CVs reported for hydrometry (HD) and DXA. Estimates of body composition by the Bod Pod have been validated against HD and DXA. Average differences between Bod Pod and

HD estimates of percent fat ranged from -4.0% to 1.9% fat. Measurements by DXA and Bod Pod varied from -3.0% to 1.7% fat. [entire paragraph (79)]

Compared to UWW results ADP underestimated the percentage of body fat (in absolute terms) by 8% in lean female athletes, underestimated the percentage of body fat at lower fat values and overestimated at the higher fat values in boys and under-predicted the percentage of body fat by an average of 2% in male college football players. [entire paragraph (67)]

Bod Pod is a reasonable alternative to HD. UWW and ADP are limited by the validity of the assumptions that underlie conversion of  $D_b$  to composition, and when it is unclear whether the assumptions are met. HD and ADP are best used in combination with other methods in three-component and four-component models. [entire paragraph (79)]

#### Comment on the current study:

Both methods were not used in the current study, because there is currently no possibility for densitometry in Graz and Christchurch. Moreover those two procedures were too expensive and also time consuming for the current project, further it was our objective to compare methods for in-field use.

#### **4.1.2.3 Hydrometry (HD)**

Water is the largest component of the body, except in very obese subjects where the fat content exceeds the water (82). The body water content varies from 70% to 75% at birth and to 40% in obese adults (82). Body water can be used to estimate body composition on three levels: molecular, cellular and tissue (82). Total body water can be used to estimate both FM and FFM assuming a constant hydration of 72-73% (67). The volume of water in the body can readily be measured by isotope dilution using tritium, deuterium, or  $O^{18}$ -labeled water (82). With careful attention to detail, total body water can be measured with a precision and accuracy of 1% to 2% (82). Body water can be used to estimate fatness within 3% and when combined with body density, within 2%. Variation in hydration levels among subjects is the main limitation of this method for the athletic population (67).

Comment on the current study:

See Densitometrie.

**4.1.2.4 Ultrasound (US)**

In 1941 Karl Theo Dussik, an Austrian psychiatrist and neurologist presented his idea of using US as a diagnostic device in his paper: "Über die Möglichkeit hochfrequente mechanische Schwingungen als diagnostisches Hilfsmittel zu verwerten (On the possibility of using ultrasound waves as a diagnostic aid) " (83). In the paper, Dussik presented the theoretical considerations in US generation, transmission and effects, and the possibility of differentiating different body tissues by US transmission through these tissues (83). The use of US scanning as SAT thickness measurement was first introduced by Booth and colleagues in 1966 (84).

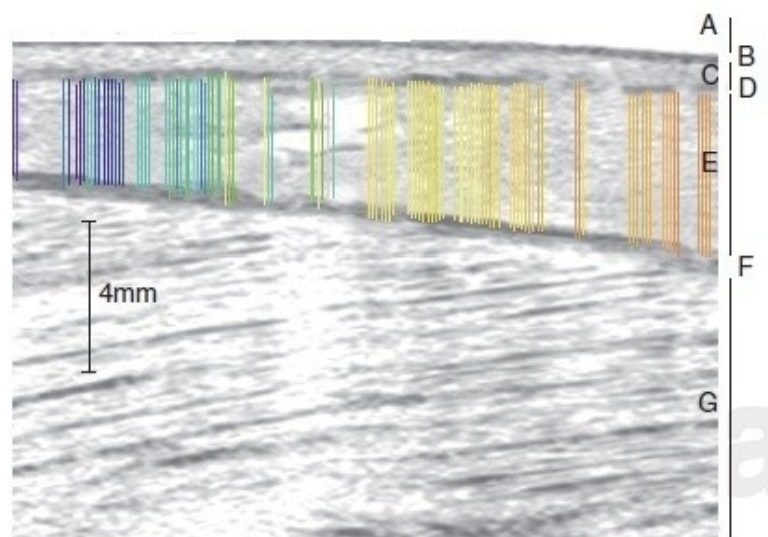
US is an oscillating sound pressure wave with a frequency greater than the upper limit of the human hearing range (> 20.000 kHz) (85). US imaging is based on the pulse-echo technique (67). Vibrations of an electrically stimulated piezoelectric crystal within a transducer produce high-frequency sound waves (1-10 MHz) (84). The beam of sound waves cross through the tissues and partially reflected from the interfaces of different underlying tissues. Soft tissues reflect only a small proportion of sound waves, while hard tissue like bones generates strong reflections (84). For 2-dimensional imaging, US beams are sent sequentially into the tissue for creating an image in which the brightness of the screen (B-mode) corresponds to the echo intensity in the plane of the scan (67). The transducer converts the echoes into electric signals that are amplified to form an enlarged image on a monitor (84).

US measurements have high precision (67, 84), but the validity of US measurements of SAT and abdominal AT is uncertain (84) with a technical error in intra- and interobserver studies was less than 0.15 mm at all sites investigated except for triceps (0.60 mm) (67, 86). Intra-observer and inter-observer coefficients of reliability for five of six ultrasonic measurements of SAT ranged from 91-98%, in comparison with coefficients of reliability ranging from 93-98% for

three skinfold measurements (86). With a standardized protocol for transducer placement and adequate operator training, B-mode ultrasonic measurements are about equal in precision to skinfold caliper measurements of SAT (84).

US has the advantage of little or no tissue compression and can be made in obese subjects and at any given sites of the body where calipers cannot be applied (e.g. sacral, paraspinal) (84).

Ultrasonic scanning is a relatively simple procedure, but interpretation of the tissue images is more difficult (67, 84), therefore a recently developed semi-automatic evaluation software can be helpful to prevent erroneous image interpretations (67) (see figure 16).



**Figure 16 Ultrasonic image after semi-automatic evaluation via a new developed software [adapted from (67)]**

#### Comment on the current study:

In the future US can become one of the most interesting alternatives compared to lipometer and anthropometry. It is able to depict zonal fat distribution with a high precision, also in lean persons as athletes, and is relative easy to transport. Nevertheless to date the access to the software for semi-automatic evaluation is limited and therefore it was no practicable alternative for the current study. However it would be a very interesting method for further evaluations.

#### 4.1.2.5 Three-Dimensional Photonic Scanning

Three-dimensional (3D) photonic scanning is a novel approach that enables the profiling of the body using structured light, class 1 (eye-safe) lasers or millimetre wave technologies (67). This digitized optical method, generating a 3D photonic image of a object, has its roots in the clothing and automotive industries and is now used to explore contrasting body shapes, for assessing body volume and percentage of fat prediction (66, 67). Currently, no study is available that assesses boy fatness in athletes via 3D scanning, because of the limited availability of scanning facilities (67).

3D scanning offers a opportunity for epidemiologic research into associations between body shape and health risks and outcome (66), and can be a advance in future body composition research in combination with other measurements as DXA (67).

#### 4.1.3 Field Methods

Field methods are selected body measurements used in field situation such as anthropometry (e.g. weight, height, circumferences, skinfolds) and bioelectric impedance analysis (BIA). Those methods are mostly less time consuming, inexpensive, non-invasive and the instruments necessary for measurements are easy to transport (84).

##### 4.1.3.1 Anthropometry

Anthropometric measurements, which describe body mass, size, shape, and level of fatness, are the most common methods of assessing body composition (3). Basic requirement for anthropometric measurements are a normal hydration status and relaxation of the muscles (68).

The **measurement of weights** is mostly done via electronic scales, which are more accurate then beam scales.

Interobserver coefficients of reliability (%) are in the range from 95% to 99% (84). The equipment has to be calibrated at regular intervals using standard weights. Growth and changes in body water, fat, and/or lean tissue leading to changes in weight. Nevertheless, body weight is highly correlated with stature (taller persons are heavier than short ones) and can therefore be misleading when taken without other measures of body size. [entire paragraph (3)].

For the **measurement of stature and sitting height** a stadiometer attached to a wall with a minimum range from 60 cm to 220 cm and including a sliding head board that is at least 6 cm wide should be used. The accuracy of measurement required is 0.1 cm. The floor should be hard and level. The stretch stature method due to International Society for the Advancement of Kinanthropometry (ISAK) criterions requires the subject to stand with the heels together and the heels,

buttock and upper part of the back touching the scale. The head, when placed in the Frankfort plane, which is achieved when the lower edge of the eye socket is in the same horizontal line as the notch superior to the tragus of the ear, need not be touching the scale. Having positioned the head in the Frankfort plane, the measurer relocates the thumbs posteriorly towards the subject's ear and far enough along the line of the jaw of the subject to ensure that upward pressure, when applied, is transferred through the mastoid processes. The subject is then instructed to take and hold a deep breath and while keeping the recorder further assists by watching that the heels do not leave the floor and that the position of the head is maintained in the Frankfort plane. Measurement is taken before the subject exhales. [entire paragraph (87)]

(Adipose) tissue distribution can be assessed with **the measurement of circumferences** (68). They are easy to perform and correlate with the body fat distribution and health risk (68). A centralized fat distribution is associated with the deposition of intra-abdominal and subcutaneous abdominal adipose tissue (3). Persons in the upper percentiles for abdominal circumference and an adverse waist/hip ratio are at increased risk for cardio vascular disease (CVD), type 2 diabetes, and mortality (see table 10) (88).

**Table 10 WHO cutoff points for waist circumference and waist/hip ratio and its risk to comorbidities (89)**

Waist circumference [cm]	Risk of comorbidities		
	increased	severe	
men	>94	>102	
women	>80	>88	
Waist-hip ratio			
	men		≥0.90
	women		≥0.85

There exist several recommendations for the placement of the measuring tape. The NIH protocol provided in the NIH Practical guide to obesity (NHLBI Obesity Education Initiative, 2000) and the protocol used in the US National Health and Nutrition Examination Survey (NHANES) III (Westat Inc, 1998) indicate that the waist circumference measurement should be made at the top of the iliac crest (88).

The NIH also provided a protocol for the measurement of waist circumference for the Multi-Ethnic Study of Atherosclerosis (MESA). This protocol indicates that the waist measurement should be made at the level of the umbilicus (88). Nevertheless published reports indicate that measurements made at the level of umbilicus may underestimate the true waist circumference (88).

Some studies have assessed the waist circumference at the point of the minimal waist (88). The WHO instructs that the measurement be made at the approximate midpoint between the lower margin of the last palpable rib and the top of the iliac crest (88). The ISAK standard for the assessment of the waist girth is the narrowest point between the lower costal (10<sup>th</sup> rib) border and the top of the iliac crest, perpendicular to the long axis of the trunk (87).

The measure should be taken at the end of the expiration phase and after an overnight fasting period to avoid affects on the accuracy of the measure (87).

The method of **skinfold measurements** relies on the assumption that there is an association between subcutaneous body fat and total body fat, however there are many factors, like age, sex and genetics that affect the subcutaneous fat mass and its distribution (68). Fatness has been predicted from skinfolds, circumferences and skeletal width, mostly validated against densitometry (67).

Currently more than 100 body fat predictions equations have been developed from skinfold measurements, leading to a flood of inconsistent outcomes due to differences in populations and lack of standardized techniques (67).

To standardize the measures and therefore to reduce the inconsistent outcomes ISAK was founded in 1986 in the UK (90). ISAK has developed international standards for anthropometric assessment and an international anthropometry accreditation scheme (IAAS) (90).

The ISAK scheme quantifies the intra- and inter-tester error, to avoid systematic differences between measurers (67). To achieve reliable data the measures should be undertaken from an trained measurer with an accurate, calibrated caliper, and a non-extensible, flexible (steel) tape (87).

Generalized equations of skinfold method provided an SEE of 5%, which is an unacceptable error, however when variation in the reference method was taken into account SEE was estimated to be less than 3% (67). While many generalized equations have been cross-validated for specific samples, their use in determining fatness in athletes relies on conforming to the assumptions both of anthropometry and densitometry (67). Only three equations were found to be reliable for use in athletes, the equation developed by Lohman with three skinfolds, Thorland et al. with seven skinfolds and Behnke and Wilmore with a combination of skinfolds, circumferences and skeletal widths; all predicted minimum weight with a total error of 2.5 kg (67). This approach involved young adult wrestlers, but could be generalized to other athletic groups if a large validation study were performed (67).

In summary, anthropometry techniques provide a simple and portable field method for estimating body composition via surrogate measures for fatness and

muscularity, they are a widespread utility for monitoring the body composition in athletes (67).

#### Comment on the current study:

Skin fold measurement provide similar advantages like the lipometer. It is easy to use, portable, inexpensive, not invasive, uses no radiation.

Compared with the skin fold method the lipometer causes only minimal pressure on the particular body site during the measurement, resulting in nearly no tissue compression. In case of the lipometer only one skin layer is between the subcutaneous fat, compared to the skin fold method, where the skin is pinched between two fingers and a double layer of skin tissue surrounds the subcutaneous fat, this can reduce measurement errors and tissue compression in case of the lipometer.

One measuring cycle at 15 body sites with the lipometer takes 2 minutes, compared to 20 to 30 minutes for skin fold measurement (landmarking, measurement, calculation), so the lipometer is timesaving, which is important in field.

Moreover the skin fold measurement predict percentage of body fat from densitometry with an SEE of 3.5%, which is not enough accurate for proper interpretation of health and performance optimization in athletes.

#### **4.1.3.2 Bioelectrical Impedance Analysis (BIA)**

BIA determines FFM and TBW in subjects without significant fluid and electrolyte abnormalities, when using appropriate population, age or pathology-specific BIA equations and established procedures (91).

The basic principle underlying the usage of BIA for determining body composition is the relation of body composition to the body water content (92). BIA is based on static assumptions and dynamic relations in terms of electrical properties of the body like its composition, hydration, density; the age, race, sex, and physical condition of those assessed (92).

BIA is based on the transfer of a low-voltage alternating current through the body (93) and the key assumptions are that the conductor is cylindrical in shape and that the current is distributed throughout the conductor uniformly (67).

Due to their water and electrolyte content lean tissue and fluids are good conductors in contrast to fat and bone (93). The principle, that the total volume of a conductor can be estimated from its length (L) and the resistance (R) to a single frequency electric current ( $L^2/R$ ), has been implemented to body composition assessment using BIA (67).

R is proportional to the length of a conductor and inversely proportional to its cross-sectional area (92). R and reactance ( $X_c$ ) are a product of the voltage drop of the applied current, they were used with height, weight, age and gender in a large number of multiple regression models to calculate body composition compartments such as FFM, lean body mass (LBM), extracellular mass (ECM), and BCM (93). R is the same as in non-biological conductors and  $X_c$  is caused by the capacitant effect of cell membranes, tissue interfaces, and non-ionic tissues (92).

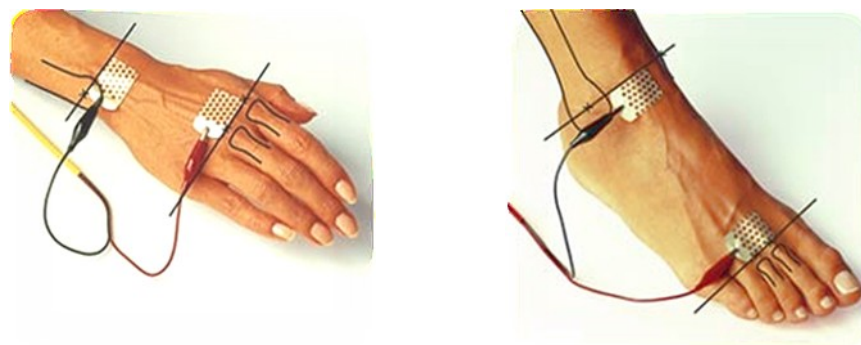
Depending on the current frequency the electric current flow differential through the extracellular- and intracellular water (ECW, ICW). At low frequency (5 kHz or less) the current flows through ECW, as the frequency increases, the current enters also the intracellular space. At frequencies above 100 kHz all body tissues were penetrated by the current. Intra- and interindividual differences in tissue composition are responsible for variation in specific resistivities among body tissues and segments and between individuals. This variation, in part, contributes to some of the interindividual differences and predictive errors in the usage of impedance to estimate body composition. [entire paragraph (92)]

The most common way is to measure the impedance is the tetrapolar method. Two electrodes are attached to the body through which the alternating electric current enters the body. Two detection electrodes are also attached to the body within the linear location of these two current electrodes, and the values of

resistance and reactance are measured across these two detection electrodes. The distance, the type and number of electrodes, and the electrode placement schemes are dependent on the device and manufacturer. [entire paragraph (92)]

Early measures of impedance were taken with the subject in a supine position and the electrode connected to the right hand-wrist and right ankle-foot (see figure 17). When newer measurements like segmental impedance and multifrequency impedance analyzers are used, measurements are taken from hand to hand or from foot to foot with the subject standing or supine. [entire paragraph (92)]

Single-frequency impedance analyzers are limited in their ability to separate total body water into intra- and extracellular compartments. The multifrequency impedance analyzer offers the ability to discriminate between ICW and ECW, this is potentially important to describe fluid shifts and balance and to explore variations in levels of hydration. [entire paragraph (92)]



**Figure 17 Electrode placement schemes [adapted from (94)]**

Even though BIA has been widely used to estimate body composition, and many equations have been reviewed, its accuracy is limited in estimating body water and body fatness. In a careful comparison between BIA and skinfolds among wrestlers, both methods predicted percentage of body fat from densitometry with an SEE of

3.5%, which is not enough accurate for proper interpretation of health and performance optimization in athletes. Additional limitations of the BIA method for athletes are the preconditions, which include abstaining from exercise. [entire paragraph (67)]

Single-frequency impedance is a useful technique for assessing TBW and FFM in groups both healthy and with disease, even though there are numerous equations for estimating these values. Impedance is a useful method of predicting body composition and is better than anthropometry alone. Currently multifrequency impedance is the area where significant gains in knowledge will occur in the next few years. [entire paragraph (92)]

Comment on the current study:

BIA is a feasible, quick field method, nevertheless BIA is not able to depict zonal fat distribution and is dependend on fluid and electrolyte abnormalities, which leads to measurement errors in athletes, and therefore BIA was not the method of choice for this study.

## 5 Discussion

Our data shows that athletes and non-athletes of both sexes can be distinguished very clearly by their subcutaneous fat patterns. In spite of comparable BMI in the males, and even significantly higher BMI in the female athlete group, the measured SAT-Top values were significantly lower in the athletes compared to non-athletes in both groups. Male and female athletes showed approximately 40-50% lower Total SAT thickness compared to non-athletes. Consequently, the results of our study illustrate that BMI is not an accurate method to evaluate fatness in young athletes and non-athletes of both sexes.

The ability of BMI to accurately reflect the amount of body fat across athletic and non-athletic populations has been assessed previously (8, 95). Nevill and colleagues (8) report a 5-32% lower total skinfold thickness (measured by callipers) in male and 5-29% lower skinfold thickness in female athletes compared to their non-athletic controls. Furthermore, when Witt and Bush (95) examined the relationship between BMI and body fat in college athletes, the authors found that only 20% of women and 4% of men with BMI  $\geq 25 \text{ kg/m}^2$  were above the 85th percentile for skinfold measurements. Ode and colleagues (9) analysed the sensitivity, specificity, and predictive values for BMI as a measure of body fatness (measured via air displacement plethysmography) and found low sensitivity between BMI and body fat percentage for athletic populations.

Other investigators have examined the diagnostic ability of BMI in relation to TBF% in adults (40, 96-102). Our data as well as results of previous researchers show that BMI is a relatively poor indicator for the amount of body fat in young athletes and non-athletes. However, because of the lack of an established TBF% criterion for health status and the differences in study design, it is difficult to compare the results of our study with this previous research. Many of these studies used different methods for measuring TBF%, including DXA (96, 97, 99), skinfolds (98) and hydrodensitometry (40, 100, 102). The different TBF% cut points used to identify over fatness included 25% (40, 97), 30% (98, 100), 33%

(102), 35% (99) and 38% (96) for females, and either 20% (40, 97) or 25% (98, 99, 102) for males. With the exception of one study that assessed postmenopausal women (96), each study assessed both males and females. The majority of studies included young, middle-aged and older adults (40, 97-100), whereas an additional study focused primarily on young and middle-aged adults (102). Within the postmenopausal women, BMI seemed to be a good diagnostic test for overfatness (96), however, the remaining research consistently indicated BMI had low sensitivity (0.06-0.60) and high specificity (0.86-1.0) as a measure of TBF% in both males and females (40, 97-100, 102).

The results of our current study suggest that BMI is not an accurate predictor of overfatness in young athletes and non-athletes, indicated by the large differences between Lipometer-determined subcutaneous adipose tissue thicknesses and BMI values. Due to a larger muscle mass among the male and female athletes, BMI incorrectly classified normal fat athletes as overfat (9, 48). Therefore, our results indicate that the subcutaneous fat patterns are a better screening tool to characterize fatness and moreover for detailed fat distribution in physically active young non-athletes. This is particularly noteworthy, given that fatness is more influenced by sport (and therefore physical training) than is the patterning of fat (103).

Our results of the ROC curve analysis showed that in men the neck body site and the trunk compartment have the highest discrimination power between the groups of athletes and non-athletes (figure 9). In women the highest discrimination power was archived at the upper back body site, and the arms compartment (figure 10). Also in previous published papers (50, 56) the neck body site became apparent as a good discriminator between normal weight healthy subjects and normal weight type-2 diabetes subjects.

The above findings confirm the dangers of using BMI in epidemiological studies, especially when a significant proportion of subjects come from a younger athletic population. When we monitoring trends in fatness over time and between populations, a more valid method of assessing fatness is likely to be obtained

using surface anthropometry such as the measurement of the neck or trunk compartment (for males) and the upper back or arms compartment for females with the Lipometer.

Other methods to assess the body composition frequently lack precision and reproducibility (calliper techniques), entail the risk of radiation exposure [computed tomography (CT), dual energy X-ray absorptiometry (DXA)], depend on hydrational status (bioimpedance), are inconvenient and time-consuming for the patient (hydrodensitometry) and/or are expensive (nuclear magnetic resonance, CT, air displacement plethysmography) (13). The Lipometer offers a new practical approach for body fat measurement.

## 6 Perspectives

We have found that the subcutaneous fat patterns are a useful screening tool for risk phenotypes in adults (50, 53, 56, 104).

Whether the subcutaneous fat patterns are also useful for assessing risky phenotypes in adolescent and physically active young people is a subject of further investigations. However, to date, there is no adequate measurement system for a rapid, inexpensive, precise, portable, and safe determination of SAT distribution. SAT-Top as measured by the Lipometer meets these criteria. Based on the good discrimination results obtained from the present dataset, Lipometer SAT-Top measurements are likely to contribute to this interesting field in further studies.

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# 9 Appendix

Datum: 23.01.2007

Name: ~~XXXXXXXXXX~~  
 Geschlecht: weiblich  
 Alter: 26.8 Jahre  
 Größe: 161 cm  
 Gewicht: 52 kg

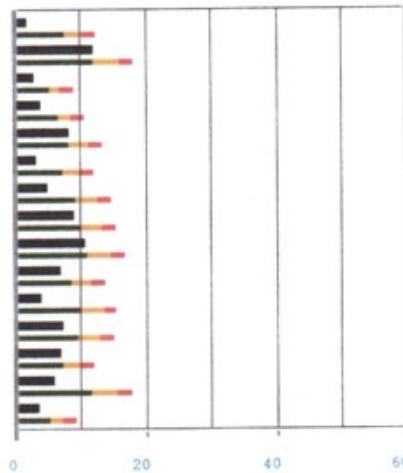
**SF-Top Protokoll**  
 gemessen vom  
**LIPOMETER V.3.0**  
 von  
**MOELLER MESSTECHNIK**

**LIPoware**  
 Subkutane  
 Fett  
 Topographie

**MESSPUNKTE**

1-Nacken	1.6	mm
2-Triceps	12.0	mm
3-Biceps	2.6	mm
4-Rücken oben	3.6	mm
5-Brust vorne	8.0	mm
6-Brust seitl.	3.2	mm
7-Bauch oben	4.7	mm
8-Bauch unten	9.0	mm
9-Rücken unten	10.6	mm
10-Hüfte	6.7	mm
11-O.Schenkel v	3.6	mm
12-O.Schenkel s	7.0	mm
13-O.Schenkel h	6.9	mm
14-O.Schenkel i	5.7	mm
15-Wade	3.3	mm

**FETTSCHICHTEN (in mm)**

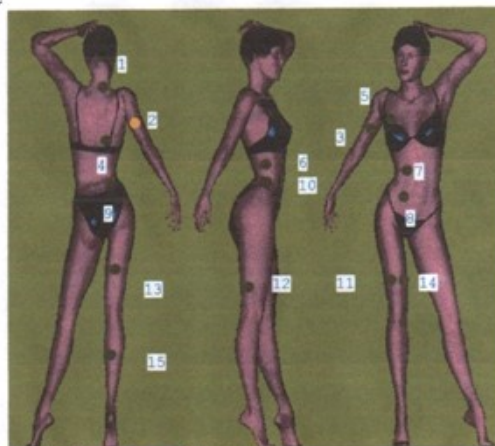
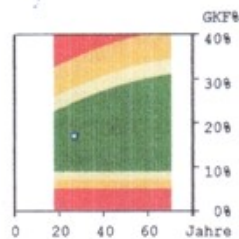


<b>GesamtKörperFett</b>	<b>17.1 %</b>	<b>8.9 kg Fettmasse</b>
sehr gut bis zu	24.7 % =	43.1 kg Magermasse
gut bis zu	28.0 % =	--- kg Fett zuviel
mittel bis zu	31.3 % =	--- kg Fett zuviel
mäßig bis zu	34.7 % =	--- kg Fett zuviel
schlecht ab	34.7 %	--- kg Fett zuviel

Subcutaneous Adipose Tissue  
 SATmass 7.8 kg  
 Visceral Adipose Tissue  
 VATmass 1.0 kg

**SF-Top: zonale Bewertung**

**GKF% - Alter**



(c) MOELLER MESSTECHNIK - Graz/Austria/Europe Email: reinhard.moeller@meduni-graz.at

Figure 18 Lipometer print out

## 10 Curriculum vitae

Renate Kruschitz, MD

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Phone: +43 1 40400 6132

Year of birth: 1980



### WORK EXPERIENCE:

- Oct. `13 – now      **Resident physician**  
Univ. Clinic for Internal Medicine, Division for Endocrinology and Metabolism, Vienna, Austria
- Aug. `13 – Oct. `13 **Resident physician**  
Clinical centre for orthopaedics and rheumatology, Bad Gastein, Austria
- Aug. `11 – Oct. `13 **Research fellow**  
Univ. Clinic for Internal Medicine, Division for Endocrinology and Metabolism, Vienna, Austria
- Jan. `11 – June `11 **Resident physician**  
Alexander Doder M.D., Graz, Austria  
Specialist for internal medicine
- Jan. `06 – June `11 **Research fellow**  
Institute of Pathophysiology of the Medical University, Graz, Austria
- July `05 – now      **Dietician on a self-employed basis**
- Sept. `02 – Aug. 03' **Dietician**  
Clinical centre for orthopaedics and rehabilitation  
Warmbad-Villach, Austria

### EDUCATION

- Okt. `10 – now      Doctoral school for lifestyle-related diseases  
Medical University, Graz, Austria
- Okt. `03 – July `10      Medical student  
Medical University, Graz, Austria  
Graduation: 13.07.2010
- Okt. `99 – Sept. `02      BS in dietetics and nutrition  
Academy for dietetics and nutrition, General Hospital, Vienna, Austria
- Finished 1999      High school diploma  
CHS – Centrum Humanberuflicher Schulen, Villach, Austria

**COURSES / OTHERS**

July 2012	Good Clinical Practice ( <b>GCP</b> ) Training, Meduni Wien, Österreich
Feb. 2011	<b>ISAK</b> (Internat. Society for the Advancement of Kinanthropometry) Level One Anthropometrist, Graz, Österreich
2005 - 2008	Singing education, Carolina Astanei, opera singer, Graz, Austria
Feb. 2007	Erasmus Intensive Programme Combating Obesity: Strategies for Prevention and Intervention, Graz
Nov. 2002	Diabetes schooling according to the Carinthian model Medical association of Carinthia, Klagenfurt, Austria
Feb. 1998	Schooling for Paramedic Red Cross, Villach, Austria

**COMPUTER SKILLS:** windows applications, EWP, SPSS

**LANGUAGES:**

German:	Mother tongue
English:	advanced
Italian:	basics

**AWARDS:**

- 2009 **GESKES-award** of the swiss society for clinical nutrition, within the framework of the 8th NUTRITION Congress, 4.-6. June 2009, Zürich, Switzerland.
- 2003 **Milupa-award** for dietetics and nutrition, within the framework of the 20<sup>th</sup> congress of the Austrian dieticians 13.-14. March 2003, Vienna, Austria

**MEMBERSHIPS:**

Arbeitsgemeinschaft für klinische Ernährung (AKE)  
The European Society for Clinical Nutrition and Metabolism (ESPEN)  
American College of Sports Medicine (ACSM)