## Body Composition in Athletes:

## FROM METHODOLOGY TO APPLICATION

## Dissertação elaborada com vista à obtenção do grau de Doutor em <br> Motricidade Humana na especialidade de Atividade Física e Saúde

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## Abbreviations

| $\sum$ 7SKF | Sum of seven skinfolds (triceps, subscapular, biceps, suprailiac, <br> abdominal, thigh, and medial calf) |
| :--- | :--- |
| $\sum_{\text {app }}$ SKF | sum of appendicular skinfolds (triceps, biceps, thigh, and medial calf) |
| $\sum_{\text {arm }}$ SKF | sum of arm skinfolds (triceps and biceps) |
| $\sum_{\text {legSKF }}$ | sum of leg skinfolds (thigh and medial calf) |
| $\sum_{\text {trunk }}$ SKF | sum of trunk skinfolds (subscapular, suprailiac, and abdominal) |
| ADP | air displacement plethysmography |
| AEE | activity energy expenditure |
| ALST | appendicular lean soft tissue |
| BCM | body cell mass |
| BIA | bioimpedance analysis |
| BIS | bioelectrical impedance spectroscopy |
| BM | body mass |
| BMD | bone mineral density |
| BV | body volume |
| CT | computed tomography |
| Db | body density |
| DIT | diet-induced thermogenesis |
| DLW | doubly labelled water |
| DXA | Dual Energy X-ray Absorptiometry |
| ECS | extracellular fluid |
| ECW | extracellular water |
| energy expenditure |  |


| EI | energy intake |
| :--- | :--- |
| FFM | fat-free mass |
| FFM $_{\mathbf{D}}$ | fat-free mass density |
| FM | fat mass |
| HR | heart rate |
| ICF | intracellular fluid |
| ICW | intracellular water |
| LST | lean soft tissue |
| Mc | muscle circumefrences |
| Mo | bone mineral |
| MRI | magnetic resonance imaging |
| Ms | soft-tissue mineral |
| PAL | physical activity level |
| REE | resting energy expenditure |
| SM | skeletal muscle mass |
| TBK | total body potassium |
| TBW | total body water |
| TEE | total energy expenditure |
| UWW | underwater weighting |
| VOmax | Maximal oxygen consumption |


#### Abstract

It is recognized that an accurate body composition assessment is relevant for prescribing adequate training and nutritional regimens in highly trained athletes. The present dissertation presents four research studies conducted under the scope of the body composition methodological and alteration research areas. In the methodological area, an alternative solution to evaluate participants taller than the DXA scan area was valid and simple to be used in athletes engaged in sports recognised for including very tall competitors. Another study was performed to test the validity of a combined motion sensor (accelerometer and heart rate monitor) in assessing total and activity energy expenditure of highly trained athletes. Using doubly labelled water as the reference method, the combined sensor was accurate for estimating energy expenditure at a group level but was of limited validity for assessing energy requirements in athletes. Under the scope of the research area on body composition alterations, a very detailed characterization of body composition changes at the molecular, cellular, tissue, and whole-body level of analysis in elite junior basketball players during the course of a season was studied. The season was associated with an improved body composition profile, particularly in males. Considering the relevance of an accurate body composition and energy balance regulation over the season, a last study was conducted to provide reference percentiles ( $5^{\text {th }}, 25^{\text {th }}, 50^{\text {th }}, 75^{\text {th }}$, and $95^{\text {th }}$ ) for several anthropometric and DXA body composition variables, stratified by sex and sport. These reference percentiles should be a helpful tool for coaches and nutritionists, in both laboratory and field settings, to prescribe exercise training and dietary intake regimens that assure an adequate energy requirement regulation in athletic populations over the season.


Key-words: body composition; athletes; season; dual-energy X-ray absorptiometry; dilution techniques; multi-component models; reference values; energy expenditure; doubly labelled water; physical activity monitors


#### Abstract

Resumo

É reconhecida a relevância de uma avaliação válida da composição corporal para a prescrição adequada de programas de treino e dieta alimentar em atletas de alta competição. Esta dissertação apresenta quatro trabalhos de investigação conduzidos no âmbito de duas grandes áreas de investigação da composição corporal, metodologia e alterações. No âmbito da área metodológica foi avaliada uma alternativa para determinar a composição corporal em atletas cuja estatura excede a área de scan da DXA. Esta solução mostrou-se válida e simples para avaliar atletas muito altos, normalmente envolvidos em desportos onde esta característica apresenta uma vantagem competitiva. Foi realizado um outro estudo para testar a validade de um sensor de movimento que combina acelerometria e cardiofrequencímetro na avaliação do dispêndio energético total e em atividade física de atletas de alta competição. Tendo como referência a técnica da água duplamente marcada, o sensor combinado apresentou-se como um método válido na estimação do dispêndio energético num grupo de atletas, embora na avaliação das necessidades energéticas individuais este equipamento tenha apresentado uma validade muito limitada. No âmbito da área de investigação das alterações da composição corporal foi conduzido um estudo que caracterizou de forma muito detalhada as alterações da composição corporal ao nível molecular, celular, tecidular e de corpo inteiro em jogadores de basquetebol ao longo de uma época desportiva. Foi observada uma associação entre a época desportiva e a melhoria do perfil de composição corporal, de forma mais notória nos basquetebolistas do sexo masculino. Dada a relevância de uma avaliação válida da composição corporal assim como da regulação do balanço energético ao longo de uma época desportiva, foi conduzido um último estudo que estabelece percentis de referência (percentis $5,25,50,75$ e 95 ) para diversas variáveis obtidas através de técnicas antropométricas e pela DXA, em função do género e desporto. Estes percentis de referência podem ser instrumentos muito úteis para treinadores e nutricionistas, quer a nível laboratorial como não laboratorial, de forma a prescrever um regime de treino e dieta alimentar que garanta o equilíbrio das necessidades energéticas da população atlética ao longo de uma época desportiva.


Palavras chave: composição corporal; atletas; época desportiva; densitometria radiológica de dupla energia; técnicas de diluição; modelos multi-compartimentais; valores de referencia; dispêndio energético; água duplamente marcada; equipamentos de avaliação da atividades física.

Introduction to the dissertation

### 1.1. Dissertation structure

The study of body composition in the athletic field has played an important role in monitoring athletic performance, training regimens, and also the athletic health status. The present dissertation, entitled "Body Composition in Athletes: from methodology to application" aimed to review some methodological issues relevant to the athletic field and to provide sports professionals a direction to use and apply body composition methodologies but also to understand and compare the several body components with proposed sex and sports specific references.

The present dissertation incorporates a compilation of four research articles already published, in press, or submitted for publication in peer-review journals with an established ISI Impact Factor. To clarify the framework of these studies this dissertation is organized as follows:

Chapter 2 includes a literature review of the topic, highlighting how the study of body composition is organized, particularly by looking in detail to the three body composition research areas (rules, methodology, and alterations). In addition, based on this organization, we reviewed the current literature regarding body composition along with the main gaps that currently exist regarding the study of body composition in the athletic field. This section finishes by highlighting the main research goals of the dissertation.

A detailed review of the methodology used in the present dissertation is showed in Chapter 3. Apart from the fact that in the four studies we included a methods section, we found relevant the inclusion of a methodology chapter. In this chapter we will provide a more detailed explanation of the methods used through the studies, specifically if a general description was provided.

Chapters 4 to 7 correspond to the four studies that were conducted to answer the research goals that were stated in chapter 2.

The Chapter 8 corresponds to a general discussion that provides a summary and integrated discussion of the main findings obtained within the four studies of this dissertation. This section was organized taking into account the three research areas that
were explained in Chapter 2 (literature review). Practical applications, taking in consideration the main findings, were also pointed out in the end of this section.

The bibliographic references were presented by the end of each section adopting a number format.

In the end the appendices section includes material that is mentioned across the dissertation that is essential to the integrity of the work presented.

### 1.2. List of articles and conference abstracts as first author

The investigation carried out as part of the present doctoral research program resulted in the following publications, and communications (oral/poster) as first author:

### 1.2.1. Peer-Reviewed articles published or in press that are

 related to the dissertation:Santos DA, Silva AM, Matias CN, Magalhães JP, Minderico CM, Ekelund U, Sardinha LB (in press) Validity of a combined heart rate and motion sensor for the measurement of free-living energy expenditure in very active individuals. Journal of Science and Medicine in Sports.

Santos DA, Silva AM, Matias CM, Rocha PM, Alison DB, Sardinha LB (in press). Association of an entire season with body composition in elite junior basketball players. The Journal of Sports Medicine and Physical Fitness.

Santos DA, Gobbo LA, Matias CM, Petroski EL, Gonçalves EM, Cyrino ES, Minderico CS, Sardinha, LB, Silva AM (2013). Body composition in taller individuals using DXA: A validation study for athletic and non-athletic populations. Journal of Sports Sciences. 31(4): 405-13. DOI: 10.1080/02640414.2012.734918.

Santos DA, Matias CN, Monteiro CP, Silva AM, Rocha PM, Minderico CS, Sardinha LB, Laires MJ (2011). Magnesium intake is associated with strength performance in elite basketball, handball and volleyball players. Magnesium Research. 24(4): 215-9. DOI: 10.1684/mrh.2011.0290.

Santos DA, Silva AM, Matias CN, Fields DS, Heymsfield SB, Sardinha LB (2010). Accuracy of DXA in estimating body composition changes in elite athletes using a four compartment model as the reference method. Nutrition \& Metabolism. 7: 22. DOI: 10.1186/1743-7075-7-22.

### 1.2.2 Other Peer-REVIEWED ARTICLES PUBLISHED AS FIRST AUTHOR:

Santos DA, Silva AM, Baptista F, Santos R, Mota J Sardinha LB (2012). Sedentary behavior and physical activity are independently related to functional fitness in older adults. Experimental Gerontology. 47(12): 908-12. DOI: 10.1016/j.exger.2012.07.011.

Santos DA, Silva AM, Baptista F, Gobbo LA, Mota J, Sardinha LB (2012). Are cardiorespiratory fitness and moderate-to-vigorous physical activity independently associated to overweight, obesity, and abdominal obesity in elderly? American Journal of Human Biology. 24(1): 28-34. DOI: 10.1002/ajhb. 21231.

### 1.2.3. AbSTRACTS that are related to the dissertation:

Santos DA, Silva AM, Matias CN, Sardinha LB (2011) Accuracy of a combined heart rate and motion sensor for the measurement of energy expenditure in elite junior basketball players. In Book of Abstracts of the $2^{\text {nd }}$ International conference on Recent Advances and Controversies in Measuring Energy Metabolism, Maastricht, The Netherlands, $2^{\text {nd }}$ to $4^{\text {th }}$ November 2011. p. 55

Santos DA, Silva AM, Matias CN, Rocha PM, Sardinha LB (2011). Effects Total body Water and Body Fluid Distribution Changes on Strength in Elite Basketball Players. In International Journal of Obesity, 35(Supp 2): S10-S27

Santos DA, Silva AM, Matias CN, Rocha PM, Sardinha LB (2011). Changes in FatFree Mass Composition and Density in Elite Basketball Players over an Entire Season. In International Journal of Obesity, 35(Supp 2): S10-S27

## Literature Review

### 2.1. Overview

Body composition refers to "the chemical or physical components that collectively make up an organism's mass, defined in a systematic way'" [1]. Conjecture on human body composition dates back to antiquity, about 440 B.C. with Hippocrates. By this time the Greeks believed that humans were made of the same basic elements that make up the cosmos: fire, water, air, and earth. Ingested food consisted of these elements, and digestion was thought to convert them to the four body juices, or humors: blood, phlegm, black bile, and yellow bile. Health was attributed to a balance of these four constituents of the body [2, 3]. Recently, human body composition research has become known as a distinct area of scientific investigation that studies various body components and their quantitative steady-state relations or rules. However the study of human body composition remounts more than 100 years and it is still an active area of basic science and clinical research. Almost every aspect of clinical nutrition, selected areas within many medical specialties and components of exercise science are touched on by the study of body composition [4]. Likewise, body composition plays an important role in the athletic field as it is associated with both sports performance [5-8] and health [9] of the athletic population.

The study of body composition is organized into three separate but interconnected research areas: body composition rules, body composition methodology, and body composition alterations (Figure 2.1) [4, 10].


Figure 2.1. The study of human body composition: three research areas [4]

The first area relates to body composition rules and studies the proportions of various components and their steady-state associations among five distinct levels: atomic, molecular, cellular, tissue-system, and whole body levels. The second area is body composition methodology and focus on in vivo methods of measuring various body components. Finally, the third area is the alteration in body composition caused by various influencing factors like growth, aging, nutrition, physical activity, race, sex, and several diseases $[4,10]$.

These three interacting areas of body composition research will be the basis of this chapter; first we will examine the rules behind body composition and its applications on the athletic population. After, we will describe the most commonly used methods for body composition assessment. Also we will look over body composition alterations with a particular focus on physical activity and energy expenditure as this is a major influencing factor in this research area. Finally we will include a section for body composition in athletes where we will review the investigations regarding the body composition rules, methodology, and alterations in athletes.

### 2.2. Body composition rules

With the purpose of organizing and systematizing the study of human body composition Wang et al. [4] as proposed a five-level model. In this comprehensive model body mass (BM) can be viewed as five distinct and separate but integrated levels of increasing complexity. The five levels are I, atomic; II, molecular; III, cellular; IV; tissue-system; and V, whole body (Figure 2.2).

Each of these levels is distinct, they do not overlap and the sum of all the components at each level of analysis is equivalent to body mass. An important concept when considering this five-level model is that components at higher body composition levels are composed of lower-level components. For example, adipose tissue is a tissuesystem level component, includes adipocytes at the cellular level, lipids at the molecular level, and carbon at the atomic level. [11].

Another important concept when looking at the five-level model is the existence of a body composition steady-state in which quantitative associations exist over a specified time interval between components at the same or different levels. A steady-
state or dynamic homeostasis exists during a specified time period if body mass and the mass of various components on the different levels are maintained relatively constant. The important implication of a steady-state is that there are stable proportions among the different components on the same or different levels. The steady state of body composition indicates that although there are more than 30 major components at the five levels of body composition, differing from each other, they are well organized according to determinable quantitative relations [4].


Figure 2.2. The five-levels of human body composition. ECS and ECF, extracellular solids and fluids, respectively [4]

The existence of a steady state within this first research area, the rules, allowed investigators to establish various characteristics of body components at each level of analysis and their quantitative relationships to one another within or between levels. Several commonly applied rules are that $16 \%$ of protein is nitrogen [12], $77 \%$ of fat mass (FM) is carbon [13], total body potassium $/$ body cell mass $=109.1 \mathrm{mmol} / \mathrm{kg}[14]$ or that the ration of total body water (TBW) to fat-free mass (FFM) is 0.732 [15].

In the next sections we will review the main rules that are applied in each level of the proposed model.

### 2.2.1. $\quad$ Atomic Level of Body Composition

The atomic level represents the foundation of body composition analysis and it is the starting point for the five-level approach [4].

Atoms or elements are the fundamental building blocks of the human body. About 50 of the 106 elements are found in the human body and their distributions in the various tissues and organs are well documented [16]. At this level 11 major elements are considered (equation 1) and six of these elements (oxygen, carbon, hydrogen, nitrogen, calcium, and phosphorus) account for $>98 \%$ of body mass. Oxygen alone constitutes more than $60 \%$ of total body mass in the Reference Man [16].

The equation for body mass, as defined at the atomic level of body composition is:
$B M=O+C+H+N+C a+P+S+K+N a+C l+M g+$ Residual

Where BM is body mass, O is oxygen, C is carbon, H is Hydrogen, N is nitrogen, Ca is calcium, P is phosphorous, S is sulphur, K is potassium, Na is sodium, Cl is chlorine, and Mg is magnesium.

Elements maintain relatively stable associations with other elements and with components at higher levels. The most common accepted rules in this level are: Sulphur/Nitrogen $=0.062 \mathrm{~kg} / \mathrm{kg}$, Nitrogen $/$ protein $=0.16 \mathrm{~kg} / \mathrm{kg}$; Carbon/triacyglycerol $=0.77 \mathrm{~kg} / \mathrm{kg}$; or Hydrogen $/$ body mass $=0.10 \mathrm{~kg} / \mathrm{kg}$ [11]

### 2.2.2. Molecular Level of Body Composition

The eleven principal elements described at the atomic level are incorporated into molecules that form $>100,000$ chemical compounds that can be found in the human body. Regardless it is neither useful nor possible to assess all of these chemical compounds individually in living humans, instead researchers consider chemical compounds in categories of closely related molecular species [4]

The molecular level of body composition analysis consists of five major components: water, protein, carbohydrates (glycogen), minerals (bone and soft tissue minerals), and lipid.

Table 2.1. Assumed constants of composition and density (at 360 C ) of fat, fat-free mass, and body mass [17]

| Body component | Density (g/cm3) | Fat-free mass (\%) | Reference body (\%) |
| :--- | :--- | :--- | :--- |
| Water | 0.9937 | 73.8 | 62.4 |
| Protein | 1.34 | 19.4 | 16.4 |
| Mineral | 3.038 | 6.8 | 5.9 |
| Osseous | 2.982 | 5.6 | 4.8 |
| Nonosseous | 3.317 | 1.2 | 1.1 |
| Fat-free mass | 1.100 | 100 | 84.7 |
| Fat | 0.9007 |  | 15.3 |
| Reference body | 1.064 |  | 100 |

## Water.

The most abundant chemical compound in the human body is water, which comprises about $60 \%$ of body mass in the Reference Man [16]. Water is distributed into the intracellular compartment ( $34 \%$ of BM) and the extracellular compartment ( $26 \%$ of BM) the last including five sub compartments: interstitial, plasma, connective tissue, bone, and gastrointestinal tract $[18,19]$. Water is the largest component of fat-free mass (FFM), accounting for $73.8 \%$ and its density at $36^{\circ} \mathrm{C}$ is $0.9937 \mathrm{~g} / \mathrm{cm}^{3}$ [17] (Table 2.1).

## Protein.

There are many different families of proteins but the term protein in bodycomposition research usually includes almost all compounds containing nitrogen, ranging from simple amino acids to complex nucleoproteins. The most widely used representative stoichiometry for protein is $\mathrm{C}_{100} \mathrm{H1}_{59} \mathrm{~N}_{26} \mathrm{O}_{32} \mathrm{~S}_{0.7}$ [4]. The density used for total body protein is $1.34 \mathrm{~g} / \mathrm{cm}^{3}$ and comprises $19.4 \%$ of FFM , however, specific proteins differ in density [17].

## Glycogen.

The primary storage form of carbohydrate is glycogen which is found in the cytoplasm of most cells. There is less than 1 kg of glycogen in healthy adults; the principal distribution is within skeletal muscle ( $\sim 2 \%$ wet weight) and liver ( $\sim 1 \%$ wet weight). The stoichiometry of glycogen is $\left(\mathrm{C}_{6} \mathrm{H}_{10} \mathrm{O}_{5}\right)_{\mathrm{x}}$ with an average density of 1.52 $\mathrm{g} / \mathrm{cm}^{3}[4,16,20]$.

## Mineral.

The term mineral describes a category of inorganic compounds containing an abundance of metal elements (e.g. calcium, sodium, and potassium) and non-metal elements (e.g. oxygen, phosphorus, and chlorine). The term ash is an important concept and represents the residue of a biological sample heated for a prolonged period to more than $500^{\circ} \mathrm{C}$, consisting of the non-volatile portion of mineral compounds. Total body ash is slightly lower in mass than mineral mass because of the loss of carbon dioxide from some carbonate groups and the release of tightly bound water during the heating period $[4,16,20]$. Minerals comprise $6.8 \%$ of body mass and are distributed in two main compartments: bone mineral (Mo) and soft-tissue mineral (Ms) (non-osseous) [17]. The bone mineral contains > $99 \%$ of total body calcium and about $86 \%$ of total body phosphorus [16]. Soft-tissue minerals include potassium, sodium, and chlorine [4]. The density of bone mineral is $2.982 \mathrm{~g} / \mathrm{cm}^{3}$ at $36^{\circ} \mathrm{C}$. The densities of soft-tissue minerals range from $3.07 \mathrm{~g} / \mathrm{cm}^{3}$ for potassium bicarbonate to $4.99 \mathrm{~g} / \mathrm{cm}^{3}$ for magnesium chlorine, these densities were then multiplied by their relative contributions yielding an overall density of $3.317 \mathrm{~g} / \mathrm{cm}^{3}$ for soft-tissue minerals [17].

## Lipid.

Lipids are defined as a group of chemical compounds that are insoluble in water and very soluble in organic solvents like diethyl ether, benzene, and chloroform [21, 22]. From the molecular components described above, lipids are the most confusing since the terms lipid and fat are used interchangeably.

Lipids can be divided into fat and non-fat according to distribution, function, and solubility characteristics. In an adult about $90 \%$ of total body lipids are fat. Fat consists almost entirely of triglycerides that are simple lipids. The term fat is therefore used as a synonymous of triglycerides [21, 22]. The non-fat lipids include phospholipids, sphingolipids, and steroids.

Lipids can also be classified physiologically into two groups: essential (Le) and nonessential (Ln) lipids. The essential lipids like sphingomyelin and phospholipids, serve important functions such as forming cell membranes. The nonessential lipids, largely in the form of triglycerides, provide thermal insulation and a storage depot of
mobilizable fuel. The essential lipids comprise about $10 \%$ of total body lipids while $90 \%$ are nonessential [16].

It is assumed that the density of fat at $36^{\circ} \mathrm{C}$ is $0.9007 \mathrm{~g} / \mathrm{cm}^{3}$ and this value is stable between subjects [17].

In Figure 2.3 are illustrated the several molecular components that were described above.


Figure 2.3. Molecular level components.

### 2.2.3. Cellular Level of Body Composition

The coordinated functions and interactions between cells are central to the study of human physiology. The cellular level is therefore an important area of body composition research. The cellular level of body composition is the first that includes living cells, which are the base of human physiology in health and disease but also for athletic performance. Despite its importance in the study of human body composition, little research has been conducted at this level, possibly because of the difficulty in quantifying some of the components [4].

The traditional cellular level model consists of three components: cell mass, extracellular fluid (ECF), and extracellular solids (ECS) [2, 4, 11] (equation 2).
$B M=$ cells $+E C F+E C S$

There are many relatively stable cellular-level relationships that are used in body composition research, and some of the most important are described as follows: potassium $/$ intracellular water $=152 \mathrm{mmol}$ of potassium $/ \mathrm{kg}$ of water; calcium $/ \mathrm{ECS}=$ $0.177 \mathrm{~kg} / \mathrm{kg}$; and potassium $/$ body cell mass $=109.1 \mathrm{mmol} / \mathrm{kg}[11,23]$.

## Body cell mass.

Body cell mass (BCM) can be defined as the total mass of "oxygen-exchanging, potassium-rich, glucose-oxidizing, work-performing" cells of the body [24] and corresponds to 20 to $55 \%$ of body mass in healthy adults [24, 25]. From a physiological or clinical perspective, the concept of BCM has more importance than that of the FFM. The BCM corresponds to 50 to $60 \%$ of FFM and is most likely to show the earliest effects of disease progression, medications, changes in nutrition, or physical activity level than FFM by itself $[19,26]$. The BCM is therefore a valuable "core" reference standard for energy exchange and work performance [27].

Body cell mass includes water (intracellular), protein, and minerals in all cell types, and water is the largest chemical component of body cell mass [24, 28]. The "typical" mammalian cell contains $70 \%$ water, $18 \%$ protein, $5 \%$ phospholipids, $1 \%$ inorganic ions (e.g., $\mathrm{K}^{+}, \mathrm{Na}^{+}, \mathrm{Mg}^{2+}, \mathrm{Cl}^{-}$), $1.35 \% \mathrm{RNA}$ and DNA, $2 \%$ polysaccharides, and $3 \%$ miscellaneous small metabolites [29]. Therefore BCM hydration (intracellular fluids) is assumed to be a mean value of 0.70 [2].

## Extracellular fluid.

Extracellular fluid is a nonmetabolizing component that surrounds cells and provides an intermediate for gas exchange, transfer of nutrients, and excretion of metabolic end products. Extracellular fluid is distributed into two main compartments, with about one-sixth as plasma in the intravascular space and the remaining five-sixths as interstitial fluid in the extravascular space [2, 25]. Extracellular fluid consists of water, protein, and minerals $[30,31]$ and its hydration is assumed to be about 0.98 (i.e., a proportional mix of plasma and interstitial fluid) [2].

## Extracellular solids.

The extracellular solids are a non-metabolizing portion of the human body that consists of organic and inorganic chemical compounds. From a clinical perspective they
are not of much interest, as they consist mainly of bone minerals (calcium and phosphorus) and collagen, reticular, and elastic fibres. Other inorganic components are also present in extracellular solids, including bicarbonate, citrate, magnesium, and sodium [4].

### 2.2.4. Tissue-System Level of Body Composition

The human body can be organized into tissues, organs and systems, this organization corresponds to the fourth level of body composition - the tissue-system [4]. Tissues contain cells that are similar in appearance, function, and embryonic origin [32]. The main tissue-system level components are adipose-tissue, skeletal muscle, bone, visceral organs, and brain [11]. Altogether the adipose, muscular, and bone tissues comprise approximately 75\% of the Reference Man's body mass [16].

## Adipose tissue.

Adipose tissue is a type of connective tissue made up of adipocytes with collagenous and elastic fibers, fibroblasts, and capillaries. Adipose tissue can be divided into four types according to its distribution: subcutaneous, visceral (i.e. loosely surrounds organs and viscera), interstitial (i.e. intimately interspersed among the cells of organs), and yellow marrow [16]. Additionally it is now recognized a distinction between white and brown adipose tissue. The traditional role attributed to white adipose tissue is energy storage, with fatty acids being released when fuel is required [33]. Brown adipose tissue is a specialized tissue for thermogenesis in mammals, and it has been considered as a heating system in the body for burning excess calories. The function of brown adipose tissue is to dissipate large amounts of chemical/food energy as heat, thus maintaining the energy balance of the whole body [34].

## Muscular tissue.

Muscle tissue can be subdivided into striated skeletal, smooth, and cardiac tissues. The skeletal muscle is also known as voluntary or striated, representing about $30 \%$ to $40 \%$ of body mass [16]. The majority of the skeletal muscle mass is found in the legs, with lesser amounts in the head, trunk, and arms [35].

## Bone tissue.

Bone is a specialized form of connective tissue that consists of bone cells surrounded by a matrix of fibbers and ground substance. The distinguishing feature of bone is that the ground substance is calcified and accounts for nearly $65 \%$ of dry bone mass [16]. The calcified ground substance is mainly hydroxyapatite and a small amount of calcium carbonate [36].

### 2.2.5. Whole Body Level of Body Composition

The fifth level of body composition is the whole body level and it concerns body size, shape, and physical characteristics. Both humans and some primates have similar body compositions at the atomic, molecular, cellular, and tissue-system levels. The complex characteristics that distinguish humans from all other primates are found at the whole body level of body composition. There are $\geq 10$ suggested dimensions at the whole body level. Examples of commonly used measures at this level of analysis are height, body mass, body mass index, segment lengths, body breaths, circumferences, skinfold thickness, body surface area, body volume, or body density. Changes in the whole body level of analysis are related with body composition changes in the other four levels, therefore whole body level components are often used to estimate components of the other levels of body composition [3, 4].

### 2.3. Body composition methodology

Body composition methodology is an area of investigation dedicated to the study and application of methods used to quantify components at the five body-composition levels [4]. Considering the lack of in vivo methods for body composition assessment, cadaver autopsy was the only process to acquire quantitative data on human body composition. The chemical analysis of tissues and fluids taken from the body, date from the mid- $19^{\text {th }}$ century $[17,37,38]$. As the original method of quantitative body composition research, cadaver study has had great importance even to the present day. The largest scale cadaver dissection was the Brussels study [38], in which 12 male and 13 female cadavers were dissected, accumulating considerable quantitative body composition data. It was in the mid $-20^{\text {th }}$ century, with the arrival of nuclear in vivo
chemistry direct (nondestructive, noninvasive) that chemical assays of the living human body became possible [10].


Figure 2.4. Classification of in vivo body composition methods [18]
All in vivo human body composition methods can be summarized by a basic formula, $\mathrm{C}=f(\mathrm{Q})$ (figure 2.4). The first part is the measurable quantity $(\mathrm{Q})$, in which there are two main categories of measurable quantities (property and component) and a third combined category. Therefore, most body composition methods can be organized as property-based and component-based methods. In addition, combined methods also exist in which both properties and components are used as the measurable quantities. The second part of the basic formula is a mathematical function $(f)$, than can be referred to as type I and type II. Type I methods share in common mathematical functions derived by statistical analysis of experimental observations. In contrast, type II methods share in common mathematical functions, which are developed, based on well-established models within and between individuals [18].

Body composition assessment is a valuable tool that can help coaches and sports scientists assess and monitor the success of training programs [39, 40]. The choice of a body composition method often depends on the intended purpose for which data are to be used and also on the availability of the techniques. Considering high performance sports, body composition assessment can be used to determine the effectiveness of exercise training and also to monitor the health status of the athlete [41]. Nonetheless, estimates of the effects of training on body composition are diverse, in part because different assessment techniques of varying accuracy and precision are used to quantify
exercise-related changes in body composition [42]. Many of the in vivo body composition methods rely on assumptions that may not be valid in athletes. On the other hand reference methods are often time consuming, expensive, and may expose athletes to unnecessary radiation. Bellow we will describe the most commonly used methods to assess body composition in each of the five levels [41, 42].

### 2.3.1. $\quad$ Atomic Level of Body Composition

Elemental analysis of humans is traditionally carried out in cadavers or in biopsy specimens from selected tissues and organs. Nonetheless, the main elements of the human body can now be measured in vivo by one or more methods [11]. There are several nuclear based techniques that can be used to obtain direct in vivo chemical assays of the whole body of humans, particularly the body's content of potassium, calcium, phosphorus, sodium, chlorine, nitrogen, hydrogen, and carbon can be measured with high precision and accuracy [26]. Total body potassium (TBK) can be measured by whole body counting, sodium, chlorine and calcium by delayed $-\gamma$ neutron activation analysis [24, 43], nitrogen by prompt- $\gamma$ neutron activation analysis [20, 24, 43], and carbon by inelastic neutron scattering [13, 43].

### 2.3.2. Molecular Level of Body Composition

## Molecular models of body composition

Traditionally body composition at the molecular level of analysis was studied as the sum of two compartments, where the body mass equals the sum of FM and FFM [15, 17, 44, 45]. However, at the molecular level FFM can be partitioned into several molecular components, including water, mineral, and protein [4].

Many stable relationships are recognized at the molecular level. These associations are integral to the body composition methodology area. The physical density of the molecular components is of extreme importance for methodological advances. The calculated and assumed constant densities of combined molecular level components are the basis of -two, -three, and -four molecular components level models [11]. In Table 2.2 are described some of the most commonly used 2- 3- and 4component models.

Table 2.2. Examples of body composition molecular models to estimate fat mass (kg)

|  | Author | Equation for FM (kg) estimation | Main assumptions | Methods and measures |
| :---: | :---: | :---: | :---: | :---: |
| $\begin{aligned} & \text { E } \\ & \sum_{2} \\ & 0 \\ & \sum_{n}^{N} \\ & 0 \\ & \text { N } \end{aligned}$ | Siri 1961 | $4.95 \times$ BV - $4.50 \times$ BM ${ }^{*}$ | $\mathrm{FFM}_{\mathrm{D}}=1.10 \mathrm{~g} / \mathrm{cm}^{3}$ <br> Constant proportions | UWW/ADP: BV |
|  | Behnke et al. 1942 <br> Brozek et al. 1963 | $4.57 \times$ BV $-4.142 \times$ BM | of water, protein, and mineral in FFM |  |
|  | Pace \& Rathbun 1945 | BM - $1.3661 \times$ TBW | TBW/FFM $=0.732$ | Isotope dilution: TBW |
| $\begin{aligned} & \text { E } \\ & \text { 己 } \\ & 0 \\ & \text { D } \\ & \text { O} \\ & \text { m } \end{aligned}$ | Siri 1961 | $2.057 \times \mathrm{BV}-0.786 \times \mathrm{TBW}-1.286 \times \mathrm{BM}^{*}$ | $\begin{aligned} & \mathrm{M} / \text { Prot }=0.351 \\ & \mathrm{M} \text { to Prot }=1.565 \end{aligned}$ | Isotope dilution: TBW UWW/ADP: BV |
|  | Withers et al. 1998 | $2.115 \times$ BV $-0.78 \times$ TBW - $1.348 \times$ BM | $\begin{aligned} & \mathrm{M} / \text { Prot }=0.354 \\ & (\mathrm{M}+\text { Prot })_{D}=1.569 \end{aligned}$ |  |
|  | Lohman, 1986 | $6.386 \times$ BV $+3.961 \times \mathrm{M}-6.09 \times \mathrm{BM}$ | $\begin{aligned} & \text { TBW } / \text { protein }=3.80 \\ & (\mathrm{TBW}+\text { Prot })_{D}=1.0486 \end{aligned}$ | UWW/ADP: BV DXA: M |
| $\begin{aligned} & \text { E } \\ & \text { 己 } \\ & 0 \\ & \sum \\ & 0 \\ & 0 \\ & \hline \end{aligned}$ | Selinger, 1977 | $2.747 \times$ BV - $0.714 \times$ TBW $+1.129 \times \mathrm{Mo}-2.037 \times \mathrm{BM}$ | $\mathrm{Ms}=0.0105 \times \mathrm{BM}$ | Isotope dilution: TBW |
|  | Heymsfield et al. <br> 1990 | $2.748 \times$ BV $-0.6744 \times$ TBW + $1.4746 \times$ TBBA $-2.051 \times$ BM | $\mathrm{Ms}=\mathrm{TBBA} \times 0.235$ | UWW/ADP: BV DXA: M / Mo / TBBA |
|  | Baumgartner et al. $1991$ | $2.747 \times$ BV $-0.7175 \times$ TBW $+1.148 \times \mathrm{M}-2.05 \times \mathrm{BM}$ | $\mathrm{Ms}=0.235 \times \mathrm{Mo}$ |  |
|  | Fuller at al. 1992 | $2.747 \times$ BV $-0.710 \times$ TBW $+1.460 \times$ TBBA $-2.05 \times$ BM | $\mathrm{Ms}=\mathrm{TBBA} \times 0.23048$ |  |
|  | Friedl et al. 1992 | $2.559 \times$ BV $-0.734 \times$ TBW $+0.983 \times \mathrm{Mo}-1.841 \times \mathrm{BM}$ | $\begin{aligned} & \operatorname{Res}_{\mathrm{D}}=1.39 \\ & (\operatorname{Res}=\operatorname{Prot}+\mathrm{Ms}+\mathrm{G}) \end{aligned}$ |  |
|  | Withers et al. $1992$ | $2.513 \times$ BV $-0.739 \times$ TBW $+0.947 \times \mathrm{Mo}-1.790 \times \mathrm{BM}$ | $\begin{aligned} & \operatorname{Res}_{D}=1.404 \\ & (\operatorname{Res}=\operatorname{Prot}+\mathrm{Ms}+\mathrm{G}) \end{aligned}$ |  |
|  | Siconolfi et al. 1995 | $2.7474 \times$ BV $-0.7145 \times$ CBW $+1.1457 \times \mathrm{M}-2.0503 \times \mathrm{BM}^{\#}$ | $\mathrm{M}=$ TBBA/ 0.824 |  |
|  | Wang et al. 2002 | $2.748 \times \mathrm{BV}-0.699 \times \mathrm{TBW}+1.129 \times \mathrm{Mo}-2.051 \times \mathrm{BM}$ | $\mathrm{Ms}=0.0129 \times \mathrm{TBW}$ |  |

Abbreviations: FM, fat-mass (kg); BV, body volume; BM, body mass; TBW, total body water; M, total mineral; Mo, bone mineral; TBBA, total body bone mineral; FFM $_{D}$, fat-free mass density; FFM, fat-free mass; Prot, protein; $\mathrm{M} / \operatorname{Prot}_{\mathrm{D}}$, total mineral + protein density; $\mathrm{TBW}^{2}$ Prot $_{\mathrm{D}}$, total body water + protein density; Ms, soft mineral; Res ${ }_{\mathrm{D}}$, residual density; UWW, underwater weighting; ADP, air-displacement plethysmography; DXA, dual-energy X-ray absorptiometry.
*This model was obtained considering the density of fat and fat-free mass at $37{ }^{\circ} \mathrm{C}$; ${ }^{\text {\# }}$ This model was developed considering 3.037 as the density of total mineral.

The basic 2-component models lie on the premise that the body can be divided into two chemically distinct compartments, FM and FFM, with FFM corresponding to all the remaining tissues together [46]. In these models it is assumed that the density of FM and FFM are $0.9007 \mathrm{~g} / \mathrm{cm}^{3}$ and $1.100 \mathrm{~g} / \mathrm{cm}^{3}$ [17], and also that the $\mathrm{FFM} / \mathrm{TBW}=$ 0.732 [28]. The majority of the errors associated with 2-component models falls not in the technical accuracy of the measurements but in the validity of the assumptions on composition and density of FFM, which are based on analyses of just three male cadavers [17]. Generally two component models involve the determination of body density (densitometric models) or TBW (hydrometric models) [46]. Science as
progressed and models to estimate body composition that partition body mass up to six components are now available (Table 2.2). By including more and different measured properties or other components than 2-component models, these methods typically account for more biological variability [46, 47].

## Densitometric models

The Ancient Greek civilization contribution to the study of body composition occurred when Archimedes (c.287-212 B.C.) observed that the buoyant force on a submerged object equals the body mass of the water it displaces, enabling the calculation of its specific gravity. He thus pioneered densitometry correctly observing that King Hiero's crown was in fact an alloy which included cheaper and less dense metals and was not pure gold [48]. These findings would be of extreme usefulness in 1942 when Albert Behnke [44], refined underwater weighting to estimate body density.

The density of an object is defined as its mass per unit volume; therefore if we are able to determine a person's body mass and volume we are able to calculate its density (body mass / volume). The body density ( Db ) is then usually transformed into FM using the Brozek et al. equation [17] (Table 2.1). At this regard the Siri equation [45] is also used to estimate FM (Table 2.1). However this last equation uses a value of $0.9000 \mathrm{~g} / \mathrm{cm}^{3}$ for the density of fat as the author considered a body temperature of $37^{\circ} \mathrm{C}$. Although body core temperature approximates $37^{\circ} \mathrm{C}$, the average body temperature under basal conditions in a comfortable environment is $1-2^{\circ} \mathrm{C}$ lower [49]. Accordingly, as Brozek et al. [17] used $0.9007 \mathrm{~g} / \mathrm{cm}^{3}$ for the density of fat at $36^{\circ} \mathrm{C}$ it seems more accurate to use this authors equation [50]. The two-component densitometric model will yield incorrect values for $\%$ FM if the overall density of the FFM components is different than $1.100 \mathrm{~g} / \mathrm{cm}^{3}[50]$.

There are currently two methods to estimate body density: underwater weighting (UWW) and air displacement plethysmography (ADP).

The most traditional method for determining body density is UWW. The method requires the subject to be completely submerged in water [44]. The volume of water displaced and/or the subject's body mass underwater, combined with the subject's laboratory body mass, are used to calculate the whole body density. The main
limitations and restrictions of this method are associated with the estimates of body volume (BV) and the residual lung volume [45, 51-53].

More recently the UWW technique started to be replaced by ADP, where the subject is immersed not in water but in a close air-filled chamber. Air displacement plethysmography systems consist of a single structure that contains two chambers: the front chamber is where the participant is tested while the rear chamber is where the instrumentation is housed and serves a reference volume. The system determines body density through an air displacement method. A volume perturbing element (movable diaphragm) is mounted on the common wall separating the front and the rear chambers. When this diaphragm is oscillated under computed control, it produces complementary volume perturbations in the two chambers (equal in magnitude but opposite in sign). These volume perturbations produce very small pressure fluctuations that are analyzed to yield chamber air volume. The classic relationship of pressure versus volume, at a fixed temperature, is used to solve for the volume of the subject chamber [54].

Densitometric methods allow estimation of FM using 2-component models [17, 44,45 ] but estimations of body volume are also necessary in multi-component models [47].

## Hydrometric models

Water is the most abundant constituent of the body [24, 37, 55]. No other method applied in vivo can provide FM estimates in such a wide range of mammals, from the mouse to the elephant, which differ in body mass by a factor of $10^{5}[2,56,57]$. Unlike the other molecular body components, the water compartment consists of a single molecular species $\left(\mathrm{H}_{2} \mathrm{O}\right)$, which simplifies the task of its measurement. Therefore, TBW is a common method for the assessment of body composition at the molecular level. The principle behind hydrometric models is that lipids are hydrophobic and thus free of water, which is therefore restricted to the FFM compartment. The calculation of FFM from TBW depends on the assumption of a constant hydration of FFM [58]. Pace and Rathbun [15] have reviewed chemical analytical data from several mammal species and observed that the $\mathrm{FFM} / \mathrm{TBW}=0.732$. By considering that BM equals the sum of FM and FFM it is possible to derivate that $\mathrm{FM}=\mathrm{BM}-(\mathrm{TBW} / 0.732)$ (equivalent to $\mathrm{FM}=\mathrm{BM}-1.3661 \times \mathrm{TBW}$ ). An exception to constant hydration of FFM
occurs in infancy, with higher TBW/FFM comparing to adult values, which implicates that fat-free mass density is lower in pediatric ages [59].

Total body water can be measured by isotope dilution [58]. The basic principle of dilution techniques is that the volume of a compartment can be defined as the ratio of the dose of a tracer, administered orally or intravenously, to its concentration in the water space within a short time after the dose is administered. Usually, two samples of the same fluid (blood, saliva or urine) are collected, one before the administration of the dose as a baseline sample and other after waiting a sufficient amount of time for penetration of the tracer within the compartment of interest, as an enrichment sample. Inherent in any tracer dilution technique are four basic assumptions: 1) the tracer is distributed only in the extrachangeable pool; 2) it is equally distributed within this pool; 3) it is not metabolized during the equilibration time; and 4) tracer equilibration is achieved relatively rapid. Therefore, TBW can be measured by using a tracer dose of labelled water (tritium, deuterium, or 18-oxygen). Deuterium dilution is the most commonly used tracer to estimate TBW, as it is a stable isotope, simple to obtain and with small costs than tritium or 18-oxygen. Isotope enrichment analysis can be performed using infrared spectrometry, nuclear magnetic resonance, mass spectrometry, and isotope ratio mass spectrometry [43, 58].

## Multi-component models

Traditionally body composition at the molecular level of analysis was studied as the sum of two components (Table 2.2), where the body mass equals the sum of FM and FFM [17, 44, 45]. However, at the molecular level FFM can be partitioned into several molecular components, including water, mineral, and protein [4].

Multi-component models share in common their developments from simultaneous equations, which may include two or more unknown components. As a general rule, for each unknown component estimated there must be one independent equation that includes the unknown component, the known component, and/or the measurable property. At the molecular level of analysis, measurable components include TBW by isotope dilution and mineral by Dual Energy X-ray Absorptiometry (DXA). Measurable properties used in developing molecular level multi-component models include body mass and body volume by UWW or ADP [11].

Body volume estimates are used in one term of the classical densitometric 2component model [17, 44, 45] that serves as the basis for multi-component models. The addition of an estimate of TBW by isotope dilution [45, 46] or mineral by DXA [60] allows the development of 3-component models. Later investigators extended the Siri's [45] classic 3-component model to a 4-component model by adding the bone mineral content of the FFM [61-68].

The formula for the 4 -component model, which controls for biological variability in TBW, bone mineral mass, and residual can be generated using the same concept as for the two- and three-component models [49]:

$$
\begin{equation*}
\frac{1}{D b}=\frac{F M}{\mathrm{FM}_{\mathrm{D}}}+\frac{T B W}{T B W_{D}}+\frac{M o}{M o_{D}}+\frac{R e s}{R_{D}} \tag{3}
\end{equation*}
$$

Where $D b$ is body density, $F M$ is fat mass, $T B W$ is total-body water, $M o$ is bone mineral, res, is residual, and ${ }_{D}$ is density.

By assuming the densities of the molecular components it is possible to derivate the following equation (equation 4)

$$
\begin{equation*}
B V=\frac{F M}{0.9007}+\frac{T B W}{0.99371}+\frac{M o}{2.982}+\frac{R e s}{1.404} \tag{4}
\end{equation*}
$$

Where $B V$ is body volume, $F M$ is fat mass, $T B W$ is total-body water, $M o$ is bone mineral, and res, is residual.
Although multi-component models share assumed constant densities for FM, TBW, and Mo, two main strategies are applied in developing these models. In one approach the residual BM (Res) after subtracting FM, water, and bone mineral is assumed to be protein and soft tissue minerals of known densities. The other approach is to assume a combined residual mass (i.e. protein, soft tissue mineral, and other) of known density [47]. In fact, residual mass includes protein, soft tissue minerals, and glycogen. In equation 4 a value $1.404 \mathrm{~g} / \mathrm{cm}^{3}$ is assumed for the residual mass density [69]. At this point it is important to remember that the largest components of residual mass are protein (density $=1.34 \mathrm{~g} / \mathrm{cm}^{3}$ ) and glycogen (density $=1.52 \mathrm{~g} / \mathrm{cm}^{3}$ ), in addition the residual mass also includes soft tissue minerals $\left(\right.$ density $\left.=3.317 \mathrm{~g} / \mathrm{cm}^{3}\right)$.

In 2002 Wang at al. [67] has stated that the available 4-component models did not include an accurate estimation of soft tissue mineral, which is a small but important molecular level component. Soft tissue minerals consist largely of soluble minerals and
electrolytes found in the extracellular and intracellular compartments of soft tissue. Although the mass of soft tissue minerals (about 400 g ) is relatively small in adults, its contribution to body density should be considered because soft tissue minerals collectively have a higher density $\left(3.317 \mathrm{~g} / \mathrm{cm}^{3}\right)$ than do each of the other components, including fat $\left(0.9007 \mathrm{~g} / \mathrm{cm}^{3}\right)$, water $\left(0.99371 \mathrm{~g} / \mathrm{cm}^{3}\right)$, protein $\left(1.34 \mathrm{~g} / \mathrm{cm}^{3}\right)$, and bone mineral ( $2.982 \mathrm{~g} / \mathrm{cm}^{3}$ ). At this regard Wang. et al [67] as developed a new 5-component model for FM which was simplified to a 4-component model (table 2.2) by assuming that Ms can be estimated from TBW $(\mathrm{Ms}=0.0129 \times \mathrm{TBW})$.

More sophisticated -five and -six component models have also emerged [70, 71]. Besides estimations of molecular components these equations also incorporate measurements at the atomic level of body composition.

## Dual-energy X-ray absorptiometry

Single photon absorptiometry was introduced in the early nineteen sixties as a way of quantifying appendicular bone mass. Dual photon absorptiometry methods first became clinically available in the early eighties, with the most recent advanced referred to as dual-energy X-ray absorptiometry (DXA) [72]. DXA provides whole body and regional assessment of FM, FFM and, also the estimation of bone mineral that can be used in multi-component models.

The fundamental principle of DXA is the measurement of the transmission of Xrays through the body at two different energy levels, low and high (typically 40 and 70 keV ), which passes through tissues and is attenuate at rates related to its elemental composition (density and thickness of the human tissues through which they pass) (Figure 2.5). The extent to which photon energy is attenuated is a function of the initial photon energy of the X-ray beam, the mass per unit area of the absorber material, and the mass attenuation coefficient ( $\mu_{\mathrm{m}}$ ) of the absorber. When photons at two different energies (e.g. 40 and 70 keV ) are passed through an absorber, attenuation at the lower energy can be expressed as a ratio (R) to attenuation observed at the higher energy. For a homogeneous absorber, R is a function of mass attenuation coefficient and mass fraction of each component [72]. Therefore each element has a characteristic mass attenuation coefficient and an $R$ value at a given energy. For instances, bone is rich in
highly attenuating minerals ( Ca and P ), and is readily distinguished from soft tissues [72].


Figure 2.5. Fundamental principle of dual-energy X-ray absorptiometry (DXA): the DXA measures the transmission of X-rays through the body at high and low energies. The X-ray beam energy is attenuated with the passage through tissue. The DXA body composition approach assumes that humans consist of three components that are distinguishable by their X-ray attenuation properties: bone mineral, fat tissue, and lean soft tissue (LST). [73]

The DXA body composition approach assumes that human consist of three components that are distinguishable by their X-ray attenuation properties: FM, bone mineral, and LST. In theory, solving for three unknown components requires measurement at three different photon energies. However, in practice, DXA can only resolve the fractional masses of a two-component mixture. Thus, DXA first separates pixels into those with soft tissue only (FM and LST) and those with soft-tissue plus bone mineral, based on the two different photon energies. This means that in pixels with bone mineral, soft tissue is not separately analyzed and the equipment assumes the FM content of the adjacent area analyzed. [72]. Normally, $40 \%$ to $45 \%$ of the whole body
scan contains bone in addition to soft tissue thus, a systematic individual error is introduced as there might be variations in body composition between measured and non measured areas [74]. For example, the influence of arm and thorax on body composition estimation can be underrepresented due to the relatively large areas of bone in those regions [75]. This source of systematic error can be increased when tracking body composition compartments [76].

For athletes, DXA measurement presents several advantages over other laboratory methods due to its good precision, large availability, and low radiation dose [41, 73]. The progressive replacement of the original pencil-beam densitometers by fanbeam devices in the early 1990s allowed for better resolution and faster scan times, without compromising accuracy and without increasing radiation dose substantially, thus easing the burden of use for both patient and clinicians [73, 77]. However caution must be taken when using DXA on multiple occasions (perhaps no more than four times during a sports season), not only due to the cumulative radiation dose [41], but also due to the error of measurement [78], which limits the ability to detect small body composition changes over time, leading to misinterpreting data [41]

Despite DXA's accuracy, precision, reliability, high speed, and non-invasive estimates with minimal radiation exposure [73, 79, 80], DXA is not without limitations. The main limitations pointed to this method are: algorithms calculations differ between manufacturers and are not published; pencil and fan-beam densitometers differ in accuracy; and limited active scan area [41]. This last limitation particularly affects athletes involved in sports where height is a major factor of performance, such as basketball and volleyball. Considering that it may be critical to measure people taller than the DXA scan area, alternative procedures are required to allow complete whole body scans (Evans, Prior, \& Modlesky, 2005).

### 2.3.3. Cellular Level of Body Composition

## Whole-Body Counting

Potassium is found mainly within the intracellular fluid compartment (ICF) and there is a stable intracellular potassium concentration. In addition, there is also a relatively stable relationship between ICF and BCM. The measurement of TBK by
whole body counting ( ${ }^{40} \mathrm{~K}$ ) can therefore be used to derive an estimate of ICF and BCM. Whole body counting duration ranges between 10 and 15 minutes, translating into a precision in the range of $2 \%$ to $5 \%$ for adults [81].

Moore \& Boyden [27] were the first to report a ratio of TBK to BCM of 120 $\mathrm{mmol} / \mathrm{kg}$. A BCM prediction model was thus derived as $\mathrm{BCM}(\mathrm{kg})=0.00833 \times \mathrm{TBK}$ (mmol).

However, the potassium concentration of BCM is not $120 \mathrm{mmol} / \mathrm{kg}$. Wang et al. [14] reported that $\mathrm{TBK} / \mathrm{BCM}$ can be calculated from four determinants: the BCM fraction as intracellular water (ICW) (a), the potassium concentration in ICW ([K] ICW), the potassium concentration in extracellular water (ECW) ( $[\mathrm{K}]_{\mathrm{ECW}}$ ), and the ratio of ECW to ICW ( $E / I$ ). The physiological aspects and mean magnitudes of the four determinants correspond to: $a=0.70,[\mathrm{~K}]_{\mathrm{ICW}}=152 \mathrm{mmol} / \mathrm{kg}$ water, $[\mathrm{K}]_{\mathrm{ECW}}=4$ $\mathrm{mmol} / \mathrm{kg}$ water, and $E / I=0.97$. By taking into account these determinants Wang et al. [14] yielded a mean TBK/BCM ratio of $109.1 \mathrm{mmol} / \mathrm{kg}$. An improved model was therefore developed in healthy adults as $\mathrm{BCM}(\mathrm{kg})=0.0092 \times \mathrm{TBK}(\mathrm{mmol})$.

## Dilution techniques

To measure the volume of extracellular water, and subsequently calculate extracellular fluids $[\mathrm{ECW} \times(1 / 0.98)[14]]$ the basic dilution techniques are the same as the described for TBW assessment, with the exception of the tracer (sodium bromide) and body fluid collection (plasma or saliva). The dilution of bromide in the extracellular space is typically the most used technique to estimzate the ECW. There are many methods for bromide measuring, namely fluorimetry, ion chromatography, neutron activation, mass spectrometry, and beta counting for radiobromide. The analytical bromide assay in most common use is high-pressure liquid chromatography [43, 58]. The bromide dilution method for estimating ECW, and thereby ECF, is easy to carry out and relatively inexpensive [82].

In addition, estimates of TBW by isotope dilution combined with the dilution of bromide for ECW estimation will allow the assessment of intracellular fluids (ICF = TBW - ECW) [43].

## Combined dilution techniques and dual-energy X-ray absorptiometry

Given that there are only about $30{ }^{40} \mathrm{~K}$ analytical systems worldwide, and the instrument is costly to purchase and maintain [23]. Shen et al. [82] have proposed an alternative method for estimating BCM by combining extracellular water (ECW) by bromide dilution and DXA measurements (equation 5). According to the authors, once ECF is known $(\mathrm{ECF}=1 / 0.98 \times \mathrm{ECW}), \mathrm{BCM}$ can be calculated as the difference between DXA-measured LST and the sum of ECF with ECS. The ECS can be derived from DXA-measured Mo as $1.732 \times$ Mo [83]. This approach that combines DXA and bromide dilution methods, represent a more practical strategy to assess the cellular level of body composition [82].
$B C M=L S T_{D X A}-(E C F+E C S)$
Where BCM is body cell mass, LST $_{\text {DXA }}$ is lean soft tissue, ECF is extracellular fluids, and ECS is extracellular solids

### 2.3.4. Tissue-System Level of Body Composition

## Computed tomography and magnetic resonance imaging

Imaging methods like computed tomography (CT) and magnetic resonance imaging (MRI) are considered the most accurate means available for in vivo quantification of tissue-system level of body composition [84]. There are several validation studies for CT and MRI, which include phantoms and human and animal cadavers [85-92]. Taken collectively, these studies support the validity of regional and whole body CT and MRI tissue-system level estimates.

In the late 1970s, CT systems were installed in most major medical centers. Between 1979 and 1981, Heymsfield and his group reported the use of CT to measure skeletal muscle mass, visceral organ volumes, and visceral adipose tissue [85-87, 91]. In 1982, Borkan et al. [93] have reported their classic visceral adipose tissue studies with CT, and in 1986, Kvist et al. [94] published for the first time assessment of whole body adipose tissue volumes with multislice CT.

Regarding MRI systems Foster at al. [95] were the first to introduce them in body composition research by demonstrating that in cadavers MRI could distinguish between adipose tissue and skeletal muscle. However Hayes et al. [96] presented the
first characterization of subcutaneous adipose tissue distribution in human subjects using MRI. In 1991, Fowler et al. [97] obtained 28 whole body MRIs, and one year after, Ross et al. [98] reported a 41-image model for measuring adipose and adipose free tissue distribution.

The estimation of tissue-system level of body composition with MRI and CT is essentially the same. The two methods primarily differ in the manner in which the images are acquired, which has a subsequent bearing on practical considerations. While CT uses ionizing radiation the MRI is based on the interaction between hydrogen nuclei (protons), and the magnetic fields generated and controlled by the MRI system's instrumentation [84]. CT and MRI are composed of picture elements, pixels, which are usually squares of $1 \mathrm{~mm} \times 1 \mathrm{~mm}$ and have a third dimension related to slice thickness. Volume elements are referred to as voxels. Voxels have a gray scale that reflects tissue composition and provides image contrast. Component estimates by CT and MRI are expressed as volumes except if they are subsequently converted to mass units by assuming constant tissue densities (e.g. $0.92 \mathrm{~kg} / \mathrm{L}$ for adipose tissue and $1.04 \mathrm{~kg} / \mathrm{L}$ for skeletal muscle) [11].

Nowadays, both CT and MRI are widely used for regional and whole body analysis of tissue-system level components [11]. However, CT remains impractical as a routine method because radiation exposure precludes studies in children and pregnant women.

## Other indirect methods

Because DXA instruments are widely available, are relative inexpensive, and radiation exposure is minimal, they have been proposed to estimate body composition at the tissue-system level, more particularly to estimate skeletal muscle mass (SM). A relatively large fraction of total body skeletal mass is in the appendages and a high percentage of appendicular lean soft tissue (ALST) is skeletal muscle mass, thus estimation of ALST by DXA is a potentially practical and accurate method of quantifying human SM in vivo [99]. At this regard models to estimate SM from ALST measured with DXA have been developed in adults [100], and children and adolescents [101].

Another practical alternative to estimate body composition at the tissue-system level of analysis is the use of anthropometric methods. Several equations have been developed in order to estimate SM from anthropometric variables like circumferences or skinfolds [102-105]. Anthropometric variables are also widely employed to estimate fat distribution, for instances a simple measurement such as waist circumference can indicate accumulation of abdominal fat, and it is the best anthropometric predictor of the amount of visceral adipose tissue [106, 107]

Other indirect techniques are also available at this level, for example estimation of SM can be estimated from 24-h urinary creatinine excretion or from TBK and nitrogen content by neutron-activation analysis [108, 109].

### 2.3.5. Whole Body Level of Body Composition

## Anthropometry

Anthropometry can be described as "The scientific procedures and processes of acquiring surface anatomical dimensional measurements such as lengths, breadths, girths, and skinfolds of the human body by means of specialist equipment' ${ }^{[1]}$.

Anthropometry can be applied at both laboratory and field settings and provides a simple, relative inexpensive, and non-invasive method for estimating body composition [41, 110]. Overall, anthropometric variables include lengths, breadths, circumferences, skinfold thicknesses, and body mass [111, 112]. Standardized techniques to assess anthropometric parameters have been developed to guarantee accurate measurements [111-113].

The skinfold is a central anthropometric variable as it allows approximations of adipose tissue patterning [114, 115], tissue mass fractionation [103], fat distribution [116], and somatotyping [117]. To date there are more than one hundred equations that convert skinfold values to body density or FM. However when using skinfolds measurements to estimate FM there are five assumptions implicit to convert the thickness of one or more compressed double layers of skin plus subcutaneous adipose tissue into total FM. By order the assumptions are: 1) the constant compressibility of skin and subcutaneous fat; 2) the constancy of skin thickness; 3) the constancy of the fat fraction of adipose tissue; 4) the constancy of adipose tissue patterning; and 5) the constancy of internal to external fat
ratio [118, 119]. Four of these assumptions have been found not to hold true and no validity has been established for the fifth. Based on these findings, it seems unreasonable to introduce further error by transforming anthropometric values into \%FM [119]. In fact Durnin [120] has pointed out that satisfactory comparisons could be performed within and between individuals from the gross values of certain skinfolds. To facilitate the evaluation and comparison of skinfold sums, investigators should collate large amounts of skinfold data and publish these as skinfold sum norms [119].

Anthropometric technique have widespread utility for monitoring the body composition of athletes provided that the measurer is well trained and follows a standard protocol, the assumptions of the technique are acknowledge, and the data treatments are not confounded with additional sources of error like the conversion to FM or body density [41].

## Bioelectrical impedance

The ability of tissues, and therefore the whole body, to conduct an electric current has been recognized for more than a hundred years [43]. Due to their dissolved electrolytes, the aqueous tissues of the human body are the major conductors of an electric current whereas FM and bone present relatively poor conductance properties. Tissue conductivity is directly proportional to the amount of electrolyte-containing fluid present. Therefore, the main principle of the bioimpedance method is that the resistance (R) of a low-level electrical current applied to the body is inversely related to the TBW and electrolyte distribution [43, 121].

Bioimpedance analysis (BIA) measurements are performed using four electrodes (two attached at the wrist and two at the ankle). For the single-frequency measurement (typically 50 kHz ), a weak alternating current is passed through the outer pair of electrodes, while the voltage drop across the body is measured using the inner pair of electrodes from which the body's impedance $(Z)$ is derived. The result of the current passage through the body gives a value of resistance (R) and reactance (X). Impedance is a function of these two separate quantities, and is also frequency dependent. The conductive characteristics of body fluids provide the resistive component, whereas the cell membranes, acting as imperfect capacitors, contribute to a frequency-dependent reactive component. Two assumptions are necessary to convert this information into volume of total body water. The first assumption is that the body can be modelled as an
isotropic cylindrical conductor with length proportional to the participant's height. The other assumption is that the reactance term that contributes to the body's impedance is small, such that the resistance component can be considered equivalent to body impedance [43, 121].

When these assumptions are combined BIA generally expresses TBW volume as a linear function of the resistance index height ${ }^{2} / R$ (or $h e i g h t^{2} / R$, for equations with impedance index), in accordance with body mass, age, and sometimes sex. There are several equations that have been proposed to estimate TBW, mostly using the resistance index. The 50 kHz current does not penetrate completely into the cells. Therefore BIA methods cannot measure intracellular water. Nonetheless, for TBW BIA presented a reasonable accuracy in healthy subjects compared to dilution methods. However BIA may not be valid in participants with an abnormal ECW/TBW ratio [121].

To solve this methodological issue, other bioimpedance methods have been developed. Recently, multi-frequency bioimpedance methods have been developed to assess water compartments. Bioelectrical impedance spectroscopy (BIS) is programmed to perform biophysical modeling on the impedance data. The modeling procedure involves fitting the spectral data to the Cole-Cole model using non-linear curve fitting [122]. This procedure generates Cole model terms, including Re (resistance associated with the ECW); Ri (resistance associated with the ICW); Cm (cell membrane capacitance); and exponent $a$. Characteristic frequency (or the frequency at which the effects of cell membrane capacitance are maximal) is subsequently calculated. Cole model terms are then applied to equations derived from the Hanai [123] mixture theory. ECW and ICW are thus calculated individually and TBW is calculated as the sum of the two compartments. This method provided accurate results for TBW, ECW, and ICW measures compared to dilution techniques in elite athletes [124].

### 2.4. Body composition alterations

The third area of body composition focused on alterations in body composition caused by various influencing factors, including physiological or pathological conditions [10]. Aging, sex, and ethnicity, nutrition, hormonal effects, or physical activity are recognized factors that modify body composition throughout the lifespan.

### 2.4.1. AGING

Changes with age in body composition begin at the moment of the conception ending only with death and subsequent decomposition of an organism. A division can be made into three phases: growth and development, maturity, and senescence. There is substantial concern in defining normal trajectories for changes within each of these phases, since abnormalities may be associated with disease states [125].

### 2.4.2. SEX AND ETHNICITY

Biological differences between sexes influence body composition per se and also processes that affect body composition such as the rate of growth and maturation, the timing and tempo of the adolescent growth spurt and sexual maturation, body proportions and physique, among others. Sex differences in body composition are negligible in infancy and childhood and are established during the adolescent spurt and sexual maturation. Genetic and cultural heterogeneity of racial-ethnic groups should also be recognized. Although individuals are labelled as belonging to an ethnic group, there are possible variations within each category. Variation in culturally determined habits, attitudes, and behaviour patterns specific related to diet, physical activity or other aspects of lifestyle have implications on body composition. At this regard, the majority of the data are based on American samples of different ethnic groups [126].

### 2.4.3. PhYSICAL ACTIVITY AND ENERGY EXPENDITURE

Physical activity is defined as any bodily movement produced by skeletal muscles that result in energy expenditure. The term exercise was used interchangeably with physical activity. In fact, both have a number of common elements, yet, exercise and physical activity are not synonyms; exercise is a subcategory of physical activity. Exercise is physical activity that is planned, structured, repetitive, and purposive in the sense that improvement or maintenance of one or more components of physical fitness is an objective. Both physical activity and exercise have in common a resulting increase in energy expenditure (EE) [127].

Total energy expenditure (TEE) can be described as the sum of resting energy expenditure (REE), activity energy expenditure (AEE) and the diet-induced thermogenesis (DIT) [128]. Resting energy expenditure (REE) represents the minimum amount of energy required to sustain vital bodily functioning in the post-absorptive awakened state [129]. DIT is the increase in energy expenditure associated with the digestion, absorption, and storage of food and accounts for approximately $10 \%$ of TEE [130]. AEE can be further separated into exercise activity thermogenesis and nonexercise activity thermogenesis components. The non-exercise activity thermogenesis is the energy expended in all the activities that are not sleeping, eating or sports-like, which includes all occupation, leisure, sitting, standing, and ambulation [131].

Energy expenditure must equal energy intake (the sum of energy from foods, fluids, and supplement products) to achieve energy balance [132]. Energy balance is usually calculated over longer periods of time and represents the difference between energy intake (EI) and total energy expenditure. When the balance is positive it will result in weight gain, whereas if a negative balance occurs, individuals will lose weight [133]. In many sport activities athletes are often under a negative energy balance for achieving a desirable body composition profile. However, realistic goals must be set regarding dramatic changes in body composition [132], and for that accurate measurements of free-living energy expenditure are mandatory.

The methods to estimate human energy expenditure are diverse. The doubly labelled water (DLW) technique is relatively non-invasive and allows quantification of total energy expenditure over a prolonged period of time (usually one week in highly active individuals) within $10 \%$ on the individual level, and as a result it is considered the gold standard for TEE assessment under free-living conditions. The technique is based on the administration of an oral dose of two stable isotopes of water $\left({ }^{2} \mathrm{H}_{2} \mathrm{O}\right.$ and $\mathrm{H}_{2}^{18} \mathrm{O}$ ). These two isotopes are used as tracers and the slightly heavier atoms ${ }^{2} \mathrm{H}$ and ${ }^{18} \mathrm{O}$ can be measured in different body fluids (e.g. urine). The ${ }^{2} \mathrm{H}$ is lost from the body in water alone, whereas the ${ }^{18} \mathrm{O}$ is lost in water and as $\mathrm{C}^{18} \mathrm{O}_{2}$ in breath. Therefore, the differences between the two tracer excretions rates represent the $\mathrm{CO}_{2}$ production rate (figure 2.6). The more rapid the drop in ${ }^{18} \mathrm{O}$ relative to the drop in ${ }^{2} \mathrm{H}$, the higher the energy expenditure is. Along with the information of the fuel oxidized, by using the food quotient (given by dietary records), TEE can be calculated [134]. Additionally, if
we are able to assess REE, for example by using indirect calorimetry, we are capable of estimating AEE (considering that DIT is $10 \%$ of TEE) as: $\mathrm{AEE}=\mathrm{TEE}-$ REE $-0.1 \times$ TEE.


Figure 2.6. Doubly labelled water technique [134]

The main advantage of this technique is that it does not interfere with daily activities; consequently unbiased measures of a free-living situation can be obtained [133]. Therefore, the DLW technique frequently been used in highly trained athletes, given that it allows athletes to engage in their normal training regimens [135, 136]. Additionally measures can be conducted over prolonged periods allowing the estimation of daily energy expenditure under free-living conditions and in consequence estimation of individual energy requirements [134]. Regardless, analytical procedures involved in dilution techniques are time-consuming, expensive, and involve complex methods and specialized technicians, excluding its routine use for EE assessment.[137].

To avoid the limitations of the DLW technique other objective measurements of energy expenditure in free-living conditions have been developed. This methods include, motion sensors, or devices that assess physiological responses to exercise such as heart rate (HR), body heat loss, and galvanic skin response. Other devices that combine two or more of these measures have been developed to estimate energy expenditure in free living conditions. However, investigations have been conducted that reveal that the currently available objective measures of energy expenditure may not provide reliable measurements in free-living conditions. Motion sensors, are not capable of detecting upper body movements, changes in grade during walking and running, and free weight exercises [138], and evidence exists that the relation between accelerometry and physical activity intensity (PAI) is affected at higher intensities [139, 140]. HR is often used as a physiological objective variable, directly associated with oxygen
consumption [141, 142]. The main limitation of the use of HR to estimate EE is the almost flat slope of the relationship at low expenditure levels. At rest, slight movements can increase HR, while EE remains almost the same [143]. On the other hand, HR does not present a good accuracy in estimating EE of individuals with high physical activity levels [142, 144]. The estimation of EE from HR is sport-specific; it has been well documented that the type of activity and posture can influence the relationship between EE and HR and consequently affect the estimation of EE from HR [143]. Even electronic devices that combine different objective measures have been shown to provide inaccurate estimates of energy expenditure, particularly when estimating individuals with high physical activity levels [145, 146]. Thus, it remains a continuing goal to develop and evaluate methods to estimate energy expenditure that are also affordable and minimally invasive.

### 2.5. Body composition in athletes

Assessing body composition has played an important role in monitoring athletic performance and training regimens [41]. Several discussions of body composition in athletes focus on relative fatness due to its potentially negative impact on performance [42]. However, other body components have been investigated in the past years as being determinant to sports performance. Numerous studies developed with athletic populations, have reported that an enhanced body composition might have a positive impact on performance parameters like maximal oxygen consumption [6], the onset of blood lactate accumulation [6], maximal strength [5, 8], and muscle power [5, 7].

On the other hand, some sports dictate athletes to make changes in body mass and composition that may not be the best option for an individual athlete [132]. Weightsensitive sports can be summarized in three categories: gravitational sports, in which mass restricts performance due to mechanical gravitational reasons (i.e. endurance sports, ski jumping, high jumping, or road cycling); weight class sports, in which unhealthy short-term mass reduction behaviour, associated with extreme dehydration, can be observed because the athletes anticipate an advantage when they are classified in a lower weight category (i.e. judo, wrestling, boxing, taekwondo, weight lifting or light-
weight rowing); and aesthetic sports, in which athletes or their coaches expect higher scores when their body mass and shape conform to a perceived body ideal, although their current body mass for health and performance is not appropriate (i.e. judge female sports of rhythmic and artistic gymnastics, figure skating, diving and synchronized swimming). In these weight-sensitive sports, concern related to athletes' health has been acknowledged as individuals experience extreme dieting, low $\% \mathrm{FM}$, frequent mass fluctuation and eating disorders [9, 41, 132, 147]. With extreme energy restrictions, losses of both muscle and FM may adversely influence an athlete's performance. Individualized assessment of an athlete's body composition and body mass or body image may be advantageous for the improvement of athletic performance. An optimal competitive body mass and composition should be determined when an athlete is healthy and performing at his or her best. Quantifying FM has been the prime focus of attention, but many coaches and scientists working with elite athletes recognize that knowledge of the amount and distribution of other body components can be as important to sports performance [41]. Methodology and equipment to perform body composition assessment must be accessible and cost-effective. Not all of the methods meet these criteria for the practitioner. In addition, athletes and coaches should know that there are errors associated with all body composition techniques and that it is not appropriate to set a specific body composition profile for an individual athlete [132].

### 2.5.1. Molecular level of body composition

The primary focus of the scientific community has been the molecular level of body composition analysis, particularly directed to FM, as excess fatness can have a negative influence on physical performance and it is often viewed by coaches and trainers as a major limiting factor in athletic achievements [42]. At this regard Malina [42] has combined data from several studies and presented estimated \%FM for athletes in numerous sports for both males (Table 2.3) and females (Table 2.4).

Table 2.3. Fat mass (\%) in samples of male athletes in several sports [adapted from [126]]

| Sport | Age (yrs) |  |  | FM (\%) |  |  | Reference |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | n | Mean | SD | Method | Mean | SD |  |
| Badminton | 7 | 24.5 | 3.6 | UWW | 12.8 | 3.1 | [148] |
| Baseball | 10 | 20.8 | 9.9 | TBW | 14.2 | 6.7 | [149] |
| Basketball | 10 | 20.9 | 1.3 | UWW | 10.5 | 3.8 | [150] |
| Basketball | 11 | 25.7 | 3.1 | UWW | 9.7 | 3.1 | [148] |
| Canoeing/kayaking | 19 | 21.1 | 7.1 | UWW | 13.0 | 2.5 | [151] |
| Cycling | 11 | 22.2 | 3.6 | UWW | 10.5 | 2.4 | [148] |
| Cycling | 11 | 21.7 | 1.7 | TBW | 13.7 | 2.3 | [152] |
| Cycling | 13 | 24.1 | 3.1 | UWW | 11.2 | 3.3 | [153] |
| Cycling | 63 | 21.9 | 3.2 | UWW | 11.8 | 3.3 | [151] |
| Field hockey | 14 | 23.7 | 3.6 | UWW | 10.3 | 4.4 | [148] |
| Football by modality |  |  |  |  |  |  |  |
| American football | 21 | 19.9 |  | ${ }^{40} \mathrm{~K}$ | 9.5 |  | [154] |
| American football | 16 | 20.3 | 0.9 | TBW | 13.8 | 6.7 | [149] |
| American football | 65 | 17-23 |  | UWW | 15.0 | 5.8 | [155] |
| Defensive back | 15 |  |  | UWW | 11.5 | 2.7 |  |
| Offensive back, receiver | 15 |  |  | UWW | 12.4 | 5.3 |  |
| Defensive lineman | 15 |  |  | UWW | 18.5 | 4.4 |  |
| Defensive linebacker | 7 |  |  | UWW | 13.4 | 4.1 |  |
| Offensive lineman | 13 |  |  | UWW | 19.1 | 7.0 |  |
| American football |  |  |  |  |  |  | [156] |
| Defensive back | 26 | 24.5 | 3.2 | UWW | 9.6 | 4.2 |  |
| Offensive back, receiver | 40 | 24.7 | 3.0 | UWW | 9.4 | 4.0 |  |
| Quarterback, kicker | 16 | 24.1 | 2.7 | UWW | 14.4 | 6.5 |  |
| Defensive lineman | 32 | 25.7 | 3.4 | UWW | 18.2 | 5.4 |  |
| Defensive linebacker | 28 | 24.2 | 2.4 | UWW | 14.0 | 4.6 |  |
| Offensive lineman | 38 | 24.7 | 3.2 | UWW | 15.6 | 3.8 |  |
| American football, blacks | 55 | 19.4 | 1.2 | UWW | 14.7 | 5.6 | [157] |
| American football, whites | 35 | 19.7 | 1.5 | UWW | 19.0 | 7.1 | [157] |
| Australian rules | 23 | 24.5 | 4.3 | UWW | 8.0 | 3.0 | [148] |
| Rugby union | 16 | 24.2 | 3.3 | UWW | 10.3 | 3.2 | [148] |
| Soccer | 9 | 24.8 | 1.9 | TBW | 6.2 | 1.9 | [158] |
| Soccer | 18 | 26.0 | - | UWW | 9.6 | - | [159] |
| Soccer | 22 | 24.5 | 3.5 | UWW | 6.9 | 3.3 | [160] |
| Soccer | 12 | 25.3 | 4.0 | UWW | 9.7 | 3.0 | [148] |
| Gymnastics | 7 | 20.3 | 0.9 | TBW | 4.6 | 3.3 | [149] |
| Gymnastics | 8 | 20.2 | 2.7 | UWW | 7.9 | 1.4 | [148] |
| Ice hockey | 27 | 24.9 | 3.6 | UWW | 9.2 | 4.6 | [161] |
| Lacrosse | 26 | 26.7 | 4.2 | UWW | 12.3 | 4.3 | [148] |
| Rowing | 8 | 24.7 | 3.2 | TBW | 7.3 | 1.3 | [158] |
| Rowing | 7 | 24.7 | 1.9 | UWW | 11.2 | 1.4 | [148] |
| Skiing | 9 | 25.9 | 2.9 | UWW | 6.3 | 1.9 | [162] |
| Skiing, cross-country | 11 | 22.8 | 1.9 | UWW | 7.2 | 1.9 | [161] |
| Skiing, cross-country | 11 | 24.0 | 4.5 | UWW | 12.3 | 4.6 | [151] |
| Speed skating | 33 | 18.4 | 2.9 | UWW | 11.2 | 2.8 | [151] |
| Speed skating | 6 | 22.2 | 4.1 | UWW | 7.4 | 2.5 | [163] |
| Squash | 9 | 22.6 | 6.8 | UWW | 11.2 | 3.7 | [148] |
| Swimming | 7 | 20.6 | 1.2 | TBW | 5.0 | 4.5 | [149] |
| Swimming | 13 | 21.8 | 2.2 | UWW | 8.5 | 2.9 | [162] |
| Swimming | 14 | 19.9 | 2.3 | TBW | 7.5 | 3.0 | [158] |
| Swimming | 39 | 19.1 | 4.5 | UWW | 12.3 | 4.6 | [151] |

Table 2.3. (cont.) Fat mass (\%) in samples of male athletes in several sports [adapted from [126]]

| Sport | Age (yrs) |  |  | FM (\%) |  |  | Reference |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | n | Mean | SD | Method | Mean | SD |  |
| Volleyball | 19 | 23.8 | 3.2 | UWW | 11.2 | 2.8 | [151] |
| Volleyball | 11 | 20.9 | 3.7 | UWW | 9.8 | 2.9 | [148] |
| Water polo | 10 | 25.8 | 4.6 | TBW | 8.8 | 2.6 | [158] |

Abbreviations: FM, fat mass; UWW, underwater weighing; TBW, total body water; ${ }^{40}$, potassium 40.

Table 2.4. Fat mass (\%) in samples of female athletes in several sports [adapted from [126]]

| Sport | Age (yrs) |  |  | FM (\%) |  |  | Reference |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | n | Mean | SD | Method | Mean | SD |  |
| Badminton | 6 | 23.0 | 5.3 | UWW | 21.0 | 2.1 | [164] |
| Basketball | 18 | 22.9 | 2.6 | UWW | 20.1 | 4.0 | [164] |
| Canoeing/kayaking | 21 | 21.2 | 3.7 | UWW | 22.2 | 4.6 | [151] |
| Field hockey | 13 | 19.8 | 1.4 | UWW | 21.3 | 7.1 | [165] |
| Field hockey | 17 | 22.6 | 2.3 | UWW | 20.2 | 6.0 | [164] |
| Field hockey | 10 | 19.8 | 1.2 | DXA | 18.3 | 2.7 | [166] |
| Gymnastics | 5 | 19.0 | 3.8 | TBW | 12.9 | 1.4 | [167] |
| Gymnastics | 44 | 19.4 | 1.1 | UWW | 15.3 | 4.0 | [168] |
| Gymnastics | 15 | 19.8 | 1.0 | DXA | 19.1 | 2.2 | [166] |
| Gymnastics, rhythmic | 7 | 20.7 | 2.7 | UWW | 15.6 | 5.1 | [151] |
| Handball, team | 17 | 23.2 | 1.9 | UWW | 19.0 | 3.7 | [151] |
| Lacrosse | 17 | 24.4 | 4.5 | UWW | 19.3 | 5.7 | [164] |
| Netball | 7 | 23.7 | 4.2 | UWW | 17.8 | 3.8 | [164] |
| Rowing | 19 | 23.6 | 3.9 | UWW | 18.4 | 3.9 | [151] |
| Rowing | 22 | 20.4 | 1.9 | DXA | 21.9 | 2.3 | [166] |
| Rowing, lightweight | 5 | 19.4 | 7.5 | UWW | 20.7 | 3.1 | [164] |
| Rowing, heavyweight | 7 | 20.5 | 3.4 | UWW | 24.2 | 4.2 | [164] |
| Skiing, cross country | 5 | 23.5 | 4.7 | UWW | 16.1 | 1.6 | [169] |
| Soccer | 10 | 24.4 | 4.5 | UWW | 20.8 | 4.7 | [170] |
| Soccer | 11 | 22.1 | 4.1 | UWW | 22.0 | 6.8 | [164] |
| Soccer | 10 | 19.8 | 0.9 | DXA | 21.8 | 2.7 | [166] |
| Softball | 14 | 22.6 | 4.0 | UWW | 19.1 | 5.0 | [164] |
| Softball | 17 | 20.4 | 1.4 | DXA | 20.9 | 3.9 | [166] |
| Speed skating | 9 | 19.7 | 3.0 | UWW | 16.5 | 4.1 | [163] |
| Squash | 6 | 27.4 | 5.6 | UWW | 16.0 | 4.9 | [164] |
| Swimming | 19 | 19.2 | 0.8 | UWW | 16.1 | 3.7 | [171] |
| Tennis | 7 | 21.3 | 0.9 | UWW | 22.4 | 2.0 | [165] |
| Volleyball | 36 | 21.7 | 2.5 | UWW | 15.8 | 4.8 | [151] |
| Volleyball | 13 | 23.0 | 2.6 | UWW | 11.7 | 3.7 | [172] |
| Volleyball | 13 | 21.5 | 0.7 | UWW | 18.3 | 3.4 | [172] |
| Volleyball | 11 | 22.8 | 3.4 | UWW | 17.0 | 3.3 | [164] |

Abbreviations: FM, fat mass; UWW, underwater weighing; TBW, total body water; ${ }^{40}$ K, potassium 40 .
The data summarized is mostly based on 2-component methods as an estimate based on other molecular models is limited. When using 2-component models body mass can be divided in FM and FFM and the density and composition of the FFM is
assumed to be constant [15, 17, 44, 45]. These rules are the cornerstones of the densitometric and hydrometric methods, variability in the density and chemical composition of the FFM is the primary factor limiting the accuracy of 2-component models for body composition estimation [45, 173, 174]. Conversely, deviations from the assumed proportions and density of the molecular components are possible with conditions that alter body composition such as aging, ethnicity, pregnancy, weight reduction, and several states of disease [47]. Also in athletes, variability on these assumptions has been observed [175-178]. Modlesky et al. [175] verified that in male weight trainers, with high musculoskeletal development, the FFM density $\left(\mathrm{FFM}_{\mathrm{D}}\right)$ was lower than the assumed $1.1 \mathrm{~g} / \mathrm{cm}^{3}$. This lower $\mathrm{FFM}_{\mathrm{D}}$ was primarily the result of higher TBW/FFM and a lower Mineral/FFM. Modlesky et al. [175] hypothesizes that the increased TBW/FFM partition was likely due to an increase in skeletal muscle mass since water comprises about $74 \%$ of SM. Similar results were reported by Withers et al. [179] for bodybuilders during a preparation for a competition. Contrarily to these authors findings, Silva et al. [176] observed that female adolescent athletes, but not males, majority post-pubescents, had a higher $\mathrm{FFM}_{\mathrm{D}}$ than the adult assumed value of $1.100 \mathrm{~g} / \mathrm{cm}^{3}$. These athletes showed a smaller water fraction and a higher protein fraction. In other investigation, Silva et al. [178] have observed that FFM/TBW in elite male judo athletes decreased from $72 \%$ to $71 \%$ from a period of weight maintenance to before a competition. This reduction was pointed out as the explanation for a $\mathrm{FFM}_{\mathrm{D}}$ increase between assessments as water presents the lowest density when compared to the other FFM components. Despite the fact that in this investigation the $\mathrm{FFM}_{\mathrm{D}}$ did not differ from the established $1.100 \mathrm{~g} / \mathrm{cm}^{3}$, in both periods the $\mathrm{FFM} / \mathrm{TBW}$ was different from the $73.2 \%$ assumed value from mammal studies. However, other investigations verified that the composition and density of FFM did not differ from the established values in athles [180-182].

The independent inclusion of TBW measurements, and bone mineral in multicomponent models, features a major advantage by controlling for much of the intersubject biological variability in FFM density and composition [49]. However, multicomponent assessment models are time consuming and require access to expensive and sophisticated technology, which often places them out of reach for practical applications in sport [41]. Therefore, using athletic populations only few investigations have
characterized body composition by using multi-component models, and the majority of the studies that used this method aimed to validate more practical field measures of body composition. In Table 2.5 are listed some investigations that assessed body composition in the athletic population using 4-component models:

Table 2.5. Investigations that characterized body composition with 4-component models in athletes.

| Sample | Sex | Reference |
| :---: | :---: | :---: |
| NCAA Division I collegiate athletes; <br> Sports: volleyball ( $n=7$ ), softball ( $n=16$ ), or track and field ( $n=6$ ) | F | [182] |
| 132 Collegiate athletes ( $M$ : $n=78$; $F: n=54$ ); <br> Sports: football, basketball, volleyball, gymnastics, swimming, and track and field teams | F, M | [183] |
| Middle- and long- distance runners (M: $\mathrm{n}=12 ; \mathrm{F}: \mathrm{n}=10$ ) | F, M | [180] |
| Judo athletes from the Portuguese national team ( $n=27$ ) | M | [78] |
| Professional water polo players ( $\mathrm{n}=10$ ) | M | [184] |
| Weight trainers ( $\mathrm{n}=14$ ) | M | [175] |
| Long distance runners ( $\mathrm{n}=10$ ) | M | [181] |
| 111 collegiate athletes; <br> Sports: football ( $n=41$ M), basketball ( $n=7 M, 1 F$ ), volleyball ( $n=5 F$ ), gymnastics ( $n=11 F$ ), swimming ( $n=10 M, 14 F$ ), and track and field ( $n=9 M$ and 13 F) | M, F | [177] |
| Adolescent athletes (M: $n=46$; $F: n=32$ ); <br> Sports: swimming, basketball, rugby, gymnastic, and judo) | M, F | [176] |
| Bodybuilders ( $\mathrm{n}=3$ ) | M | [179] |

Abbreviations: F, female; M, male
Despite the fact that at the molecular level the FM assessment has been the primary focus [42], investigations have been conducted in the past years to understand the importance of assessing other molecular body components in athletes. Quiterio et al. [185] have assessed adolescent athletes and verified that more hours per week of sports training were associated not only with lower FM but also with greater FFM components (TBW, lean, and bone mass). Other research study has observed that the level of practice is related to different body composition profiles, when comparing elite versus sub-elite female handball players [186]. The authors observed that the elite players not only had significantly lower \%FM but also higher bone mineral content than sub-elite counterparts. The same investigation has verified that elite players presented a clear tendency to accumulate more lean mass, particularly in the upper limbs. Differences in
body composition were also observed when comparing different court positions. Accordingly, it has been verified that in line with other physical fitness factors, FFM predicted female Olympic wrestling performance [187].

Hogstrom et al. [6] showed strong associations between FFM and the onset of blood lactate accumulation and maximal oxygen consumption weight adjusted thresholds among male and female cross-country skiing ( $\mathrm{r}=0.47-0.67$ ) and in female alpine-skiing ( $\mathrm{r}=0.77-0.79$ ) athletes. In another investigation Silva et al. [7] aimed to analyze the association between body composition changes, from a weight stable period to prior competition, on upper-body power in judo athletes. The authors verified that total body water changes were related to upper-body power variation ( $\mathrm{r}=0.672$ ). At this regard investigations have been conducted also to understand the impact of dehydration sports performance. For instances it has been investigated that hypohydration decreases resistance exercise performance [188] and that hypohydration can modify the hormonal and metabolic response to resistance exercise [189]. Maresh et al. [190] observed that during exercise the testosterone cortisol ratio may be altered by hydration state, therefore influencing the balance between anabolism and catabolism in response to running exercise performed at typical training intensities. In fact, the American College of Sports Medicine position stand on hydration and physical activity [191] has acknowledged that a body water deficit greater than $2 \%$ of body mass marks the level of dehydration that can adversely affect performance.

### 2.5.2. Cellular level of body composition

The cellular-level of body composition analyses often are neglected in sports research; consequently few investigations have presented data regarding components at this level. It has been investigated that athletes from several sports (soccer, judo, and water polo) present higher body cell mass than non-athletes of the same age [192]. In the same study the authors verified that the body composition profile differed among different competitive levels. In fact Andreoli et al. [192] observed that in male soccer teams the division 3 team presented lower BMC than those from division 1 and 2. The teams differed in their training regimens with the teams from division 1 and 2 presenting greater intensity workouts. In accordance, Quiterio et al. [185] have found an association between weekly training hours and greater levels of cellular body
components ( BCM and ECF ). Also investigations that relate components of the cellular level with sports performance have been conducted. The intracellular fluids are associated with power [7] and maximal strength [8] changes in elite judo athletes, with intracellular water reductions being associated with a decrease in strength and power performance. Moreover, the BCM is associated with aerobic performance in basketball players [193].

### 2.5.3. TISSUE-SYSTEM LEVEL OF BODY COMPOSITION

At the tissue-system level Midorikava et al. [194] observed that male college athletes (Olympic weightlifters, sumo wrestlers, rugby football players and swimmers) had higher skeletal muscle compared to untrained college students ( $33.0 \mathrm{~kg}, 47.7 \%$ FFM vs. 23.5 kg, $44.7 \%$ FFM). Also using MRI, Sanchis-Moysi et al. [195] verified that professional tennis was associated with marked hypertrophy of the musculus rectus abdominis ( $>58 \%$ than controls). The rectus abdominis hypertrophy was more marked in the non-dominant than in the dominant side. Similar results have been observed for soccer players (>26\% than controls) [196]. The use of MRI in athletic populations has also allowed verifying differences in organs size [194, 197-199]. Scharf et al. [197] observed that ventricular volume and mass indices were significantly higher in athletes than non-active controls. Similarly endurance athletes have increased ventricular volumes, diameters, wall mass, and wall thickness compared with non-athletes [198]. Also, Midorikawa et al. [194, 199] found that athletes presented greater liver and kidney masses than non-athletes. On the other hand, due to the elevated costs of reference methods to assess the tissue level, particularly the skeletal muscle, there is a lack of research that explored the associations between tissue-system components of body composition with sports performance.

### 2.5.4. WhOLE BODY LEVEL OF BODY COMPOSItION

At the whole body level the majority of the investigations have been conducted with anthropometric based methods. Traditionally the use of anthropometric variables in the athletic field have been used to estimate molecular components like FM [5, 200,

201] or skeletal muscle [202]. However, has described above, converting skinfold thickness to FM lies on several assumptions [119], that particularly in the athletic field may not be valid. Several investigations observed that skinfold based models are not accurate in estimating FM in athletic populations [148, 164, 177, 178]. Regardless, the use of anthropometry should not be discarded when assessing athletes' body composition. Anthropometric techniques have a widespread utility for monitoring athletes by providing a simple and highly portable method for estimating body composition in athletes via surrogate measures of fatness and muscularity [41]. Thereby, it has been proposed the use of summed skinfold thickness measure to capture a representative surface adiposity. In fact, Marfell-Jones [119] has suggested that investigators should collate the large amounts of skinfold data that have already been collected with the purpose of replacing FM prediction equations and publishing skinfold sum norms. This will allow researchers and coaches to better understand this new proposed indicator. In accordance researchers are starting to make this approach and presenting results of summed skinfolds rather than FM estimated by anthropometric equations. In Table 2.6 are presented some investigations that used this approach (sum of seven skinfolds: triceps, subscapular, biceps, suprailiac, abdominal, thigh, and medial calf), notwithstanding the fact that there are other studies that have presented this information. However, other researchers have used different sum of skinfolds in athletes, for example sum of 4 skinfolds [203], 5 skinfolds [204] 8 skinfolds [205-207], 9 skinfolds [208, 209], or even 10 skinfolds [210]. It is however important to provide a standardization at this respect in order to compare data from different investigations.

Other approach that is frequently used in athletes is the somatotype [117]. Somatotype is defined by three components: endomorphy, mesomorphy, and ectomorphy. Endomorphy expresses the relative amount of fat, mesomorphy refers to relative musculoskeletal development, and ectomorphy to body linearity. Somatotype analysis allows the demonstration of similarities and differences between groups of nonathletes and athletes participating in different modalities. Accordingly, it is important for identification of sports talents and when describing athletes body composition profiles [202, 211-215].

Table 2.6. Investigations that presented information related to the sum of seven skinfolds ( $\sum 7$ SKF).

| Sample | Age (years) | Sex | \7SKF (mm) | Reference |
| :---: | :---: | :---: | :---: | :---: |
| Basketball ( $\mathrm{n}=268$ ) | 17.1+1.0 | M | 67.5+20.6 | [216] |
| Basketball ( $\mathrm{n}=273$ ) | 16.7+1.2 | F | 95.9+24.3 |  |
| Cricketers ( $\mathrm{n}=14$ ) | $25.0 \pm 5.8$ | M | $69.7 \pm 17.4$ | [217] |
| Rugby sevens ( $\mathrm{n}=18$ ) | $21.9 \pm 2.0$ | M | $52.2 \pm 11.5$ | [218] |
| Rugby, Professional ( $\mathrm{n}=27$ ) |  |  | $47.0 \pm 60.8$ |  |
| Rugby, Semiprofessional ( $\mathrm{n}=17$ ) | $25.6 \pm 0.7$ | M | $65.3 \pm 4.9$ | [219] |
| Soccer ( $\mathrm{n}=33$ ) | $15.7 \pm 0.7$ | F | $103.1 \pm 35.2$ | [220] |
| Australian rules Football, field ( $\mathrm{n}=20$ ) | $24.7 \pm 7.7$ | M | $67.8 \pm 18.8$ | [221] |
| Australian rules Football, boundary ( $\mathrm{n}=15$ ) | $29.6 \pm 13.6$ | M | $65.6 \pm 8.8$ |  |
| Volleyball ( $\mathrm{n}=16$ ) | $18.5 \pm 1.5$ | M | $59.0 \pm 13.3$ | [222] |
|  |  |  | $51.1 \pm 68.1$ (after 2 years) |  |
| Volleyball ( $\mathrm{n}=14$ ) |  | M | $57.8 \pm 3.0$ |  |
| Volleyball ( $\mathrm{n}=20$ ) | $15.6 \pm 0.1$ | F | $69.7 \pm 1.1$ | [223] |
| Water Polo, National Squad ( $\mathrm{n}=14$ ) | $23.3 \pm 2.9$ | F | $99.4 \pm 22.2$ | [224] |
| Water Polo, National League ( $\mathrm{n}=12$ ) | $20.8 \pm 4.7$ | F | $116.4 \pm 33.9$ |  |

Abbreviations: $\sum 7$ SKF, sum of seven skinfolds (triceps, subscapular, biceps, suprailiac, abdominal, thigh, and medial calf).

Other anthropometric measures at the whole body level may include lengths, breadths, circumferences, skinfold thicknesses, or even body mass and height [111, 112]. These have also been widely used when assessing an athlete's body composition. For example, Alcaraz et al. [225] observed that grip strength was associated with girth (mesosternal, gluteus, upper thigh, medial thigh), and breadth (biacromial, femur) variables in elite trained male water polo players. Keogh et al. [226] verified that when comparing successful and less-successful powerlifters, anthropometric variables indicated that the weaker lifters had significantly smaller muscular circumferences per unit height than the stronger lifters. In team sports, it has been shown that there are anthropometric differences between players in different positions highlighting that specific morphological characteristics are necessary in team sports [186, 202, 227].

At the whole-body level also bioimpedance analysis have been use to assess body composition in athletes. This body composition methods has been used mainly to estimate components of other levels of analysis, particularly the molecular [228, 229], the cellular [192] and the tissue-system [129] level of body composition in athletes.

### 2.5.5. Seasonal variation in body composition

The role of body composition in the athletic population it is of extreme importance to analyse which changes may impact performance during the course of a season [42]. Studies commonly compare the body composition of athletes during several critical periods of the season.

Seasonal variations have been the primary focus of investigations looking over body composition changes in weight-category sports. In combat sports, athletes are subdivided into weight categories. In order to qualify for their respective weight category, many athletes undergo impressive weight changes preceding the competition [230-232]. This weight loss is usually carried out through the combined use of sauna, restriction of water intake, overtraining, and fasting [232-234]. Differences related to body composition may significantly influence fighting strategies (including technical and tactical skills) and consequently the physiologic profile of these athletes [7, 8, 235]. Follow-up studies using these weight-class sports have been conducted particularly to understand the impact of short-term weight reductions on body composition and consequently on sports performance. In wrestlers undergoing rapid weight loss a reduction in the cross sectional areas of skeletal muscle and subcutaneous fat in the trunk, assessed by MRI were observed [236]. Silva et al. [7] found that in judo athletes a significant mean reduction of 1.1 kg was observed in body mass from a period of weight stability to prior a competition but no mean changes were found in fat mass, fatfree mass, lean soft tissue (LST), total body water, and extracellular and intracellular water. On the other hand, in the same investigation the authors verified that TBW and ICW changes were related to changes in upper-body power determined in a bench press machine. In another study [201] NCAA wrestlers were tested in four occasions: 1) prior to pre-season training, 2) after pre-season training, 3 days prior to the first seasonal meet, 3) mid-season, one day prior to a meet, and 4) at the end of the season, 2 to 3 days
following the last meet. The authors observed that body mass, \%FM, and FM were lower at the first seasonal meet and in mid-season - one day prior to a meet compared to the pre-season, moreover FFM decreased from the pre-season to the first seasonal meet.

Despite the focus on weight-category sports, other studies have been conducted to understand changes that occur in body composition in the course of a season. Casajus et al. [237] assessed 15 male soccer players from the Spanish First Division at the beginning of the championship (after five weeks of training), and again at the beginning of the second round of the championship. The authors observed that in these elite soccer players the sum of six skinfolds showed a remarkable decrease ( $57.0 \pm 8.67 \mathrm{~mm}$ to 52.9 $\pm 8.61 \mathrm{~mm}$ ) in line with a decrease in relative fatness $(8.6 \pm 0.91 \% \mathrm{FM}$ to $8.2 \pm 0.91$ $\% \mathrm{FM})$. Also using skinfold measurements, for assessing 16 elite female handball players, Granados et al. [5] verified that \%FM decreased from the beginning of the first preparatory $(21.1 \pm 5.3 \%)$ period to the end of the first competitive period (19.2 $\pm$ $5.3 \%$ ). Gorostiaga et al. [238] assessed an elite male handball team during a 45 -week season using skinfold-predictive equations and showed that FFM increased from the beginning of the first preparatory period ( $80.7 \pm 8.8 \mathrm{~kg}$ ) to the beginning ( $81.8 \pm 9.4 \mathrm{~kg}$ ) and the end of the first competitive period ( $82.1 \pm 8.8 \mathrm{~kg}$ ), despite no significant changes were observed for FM during the season. Both Granados et al. [5] and Gorostiaga et al. [238] concluded that changes in \%FM correlated positively with changes in maximal strength and muscle power in male and female handball players which means that those who developed larger decreases in \%FM showed larger decreases in maximal strength (females) or muscle power (males and females) of the upper and lower extremities. Gonzalez-Rave et al. using bioelectrical-impedance analysis [229] assessed skeletal mass, FM, and FFM in elite female volleyball players. The authors performed four assessments during a competitive season: PRE (first week), POST (fourth week), POST 1 (eighth week) and POST 2 (24th week), and observed among other body composition variables, a significant decrease in \%FM of $2.08 \%$ from POST 1 to POST 2. Tavino et al. [239] evaluated 9 NCAA male basketball players using a anthropometry and verified that $\%$ FM decreased from the before ( $13.3 \pm 3.1 \%$ ) to after the pre-season $(9.8 \pm 1.9 \%)$ with an increase at the end of the season (11.7 $\pm$ $2.1 \%$ ). The author explained that a consistent and intense weight training and conditioning program led to the dramatic decrease in $\% \mathrm{FM}$ during the 5 weeks of the
pre-season and as the regular season began, the weight training program was gradually reduced and there was less emphasis on conditioning and more emphasis on strategy development. Both factors were attributed to the increase in \%FM that occurred by the end of season. Silva et al. [136] assessed body composition in 9 female and 8 male elite junior basketball players in the first week of the pre-season training period and again at the end of the in-season using DXA. In the female athletes the authors observed a decrease in \%FM, whereas an increase in FFM, LST, BMC, and ALST was observed. In the males an increase in FFM, LST, BMC, and ALST occurred but no changes were found for FM between assessments. The authors concluded that these changes in body composition were associated with an alteration in resting energy expenditure. Specifically, FFM changes explained an increased in REE. In addition, increases in regional LST, specifically at the upper limbs explained a raise in REE throughout the season. Also using DXA, Carbuhn et al. [240] have assessed female collegiate athletes from different sports (softball, basketball, volleyball, swimming, and track and field) in three periods of the season (off-season, preseason, and postseason). The authors found that changes in body composition variables occurred most often between off-season and in-season (preseason or postseason). Similarly, Meleski et al. [241] verified that in elite female swimmers decreases in body mass ( $-1.3 \pm 1.8 \mathrm{~kg}$ ), FM ( $-2.4 \pm 1.2 \mathrm{~kg}$ ) and $\% \mathrm{FM}$ $(-3.8 \pm 1.9 \%)$ and an increase in FFM ( $1.1 \pm 1.8 \mathrm{~kg}$ ) characterized the early part of the season, and these changes were generally maintained during the second part of the season.

The studies conducted over the course of a season reporting the changes in body composition focus on the molecular and whole-body levels of body composition analysis. The majority of these studies used body composition techniques of limited accuracy. Moreover, the rationale for evaluating and tracking body composition using certain protocols rather than others has never been well documented. As indicated by the above mentioned studies, research is lacking on the changes that occur during a sports season on other levels of body composition (i.e. cellular and tissue level). Moreover, scientific research is absent on the effects of certain body composition changes on athletes' physical performance during a season.

### 2.5.6. ENERGY EXPENDITURE AND ATHLETES

Athletes are more physically active than the general population and therefore they have higher daily energy expenditure. The average physical activity level (PAL), which represents the ratio of TEE and REE, lies between 1.4 and 1.7 in the general population with sedentary or light activity lifestyle [242]. From Table 2.7 it is possible to observe that PAL is typically higher in athletes, ranging from $2.0-5.3$. For individuals who regularly expend high amounts of energy on a daily basis, adequate nutrition is a primary concern. Athletes need to consume adequate energy to maintain a healthy body composition profile but also to maximize training effects. Low energy intakes can result in loss of skeletal muscle mass; menstrual dysfunction; loss of or failure to gain bone density; an increased risk of fatigue, injury, and illness; and a prolonged recovery process [132]. Energy balance is defined as dietary energy intake minus exercise energy expenditure. Meeting energy needs is a nutrition priority for athletes since optimum athletic performance is promoted by adequate energy intake [132]. With limited energy intake, FM and FFM will be used for fuel by the body. Loss of FFM results in the loss of strength and endurance, as well as compromised immune, endocrine, and musculoskeletal function [243]. Many athletes are chronically energy deficient, even though energy balance is not always the goal, as many times athletes seek to modify their body size and composition to achieve specific performance goals. Therefore, it is determinant to characterize athletes' energy expenditure in order to identify individual energy requirements in accordance to their individual goals [244]. These concerns have been more directed to weight-sensitive sports (i.e. gravitational sports, weight-class sports, and aesthetic sports) and also to female athletes [9, 147]. Regardless, it is important to estimate energy requirements for athletes from several sports, which will depend on individual factors related to the duration, frequency, and intensity of the exercise, sex, and prior nutritional status [132].

Total energy expenditure can be accurately evaluated using the doubly labelled water (DLW) method while avoiding any interference with training activities. In Table 2.7 a summary of investigations that used doubly DLW to estimate total energy expenditure in different athletic populations is presented.

Table 2.7. Summary of total and resting energy expenditure, physical activity level, and energy intakes in different sports determined by the doubly labeled water method, including energy intake if available.

| Sample | Sex | TEE, kJ/day (kcal/day) | REE, kJ/day (kcal/day) | PAL | $\mathrm{EI}, \quad \mathrm{kJ} /$ day (kcal/day) | Reference |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| elite swimmers during high volume training ( $n=5$ ) | F | $\begin{aligned} & 23400 \pm 2100 \\ & (5589 \pm 502) \end{aligned}$ | $\begin{aligned} & 7700 \pm 500 \\ & (1389 \pm 119) \end{aligned}$ | $3.0 \pm 0.2$ | $\begin{aligned} & 13100 \pm 1000 \\ & (3129 \pm 239) \end{aligned}$ | [245] |
| Elite lightweight rowers during high intensity and volume training ( $n=7$ ) | F | $\begin{aligned} & 16567 \pm 5103 \\ & (3957 \pm 1219) \end{aligned}$ | $\begin{aligned} & 5815 \pm 142 \\ & (1389 \pm 34) \end{aligned}$ | $2.9 \pm 0.9$ | $\begin{aligned} & 9270 \pm 1310 \\ & (2214 \pm 313) \end{aligned}$ | [246] |
| Elite junior basketball players at a competitive period ( $n=12$ ) | M | $\begin{aligned} & 19337 \pm 2851 \\ & (4626 \pm 682) \end{aligned}$ | $\begin{aligned} & 6558 \pm 1033 \\ & (1569 \pm 247) \end{aligned}$ | $2.9 \pm 0.5$ | $\begin{aligned} & 12101 \pm 2002 \\ & (2985 \pm 479) \end{aligned}$ | [247] |
| Elite junior basketball players at a competitive period ( $n=7$ ) | F | $\begin{aligned} & 14618 \pm 1012 \\ & (3497 \pm 242) \end{aligned}$ | $\begin{aligned} & 5476 \pm 431 \\ & (1310 \pm 103) \end{aligned}$ | $2.6 \pm 0.3$ | $\begin{aligned} & 7553 \pm 192 \\ & (1807 \pm 46) \end{aligned}$ |  |
| Elite synchronized swimmers, after competition when athletes engaged normal training regimens ( $n=9$ ) | F | $\begin{aligned} & 11500 \pm 2800 \\ & (2747 \pm 669) \end{aligned}$ | $\begin{aligned} & 5200 \pm 300 \\ & (1242 \pm 72) \end{aligned}$ | $2.2 \pm 0.4$ | $\begin{aligned} & 8900 \pm 1700 \\ & (2126 \pm 406) \end{aligned}$ | [248] |
| Professional soccer players during competitive season ( $n=7$ ) | M | $\begin{aligned} & 14800 \pm 1700 \\ & (3535 \pm 406) \end{aligned}$ | $\begin{aligned} & 7000 \pm 300 \\ & (1671 \pm 72) \end{aligned}$ | $\begin{aligned} & 2.19 \pm \\ & 0.31 \end{aligned}$ | $\begin{aligned} & 13000 \pm 3105 \\ & (2400 \pm 573) \end{aligned}$ | [135] |
| Elite Endurance runners in peak physical condition ( $n=9$ ) | M | $\begin{aligned} & 14611 \pm 1043 \\ & (3490 \pm 249) \end{aligned}$ | $\begin{aligned} & 6408 \pm 224 \\ & (1531 \pm 54) \end{aligned}$ | $2.3 \pm 0.1$ | $\begin{aligned} & 13241 \pm 1330 \\ & (3163 \pm 318) \end{aligned}$ | [249] |
| Adolescents Speed skaters living at a boarding school for young athletes at a pre-season period ( $\mathrm{n}=8$ ) | M | $\begin{aligned} & 16900 \pm 2900 \\ & (4037 \pm 693) \end{aligned}$ | $\begin{aligned} & 8400 \pm 500 \\ & (2006 \pm 119) \end{aligned}$ | $2.0 \pm 0.2$ | NR | [250] |
| Cyclists during the Tour de France, week 1 ( $n=4$ ) | M | $\begin{aligned} & 29375 \pm 991 \\ & (7016 \pm 237) \end{aligned}$ | $\begin{aligned} & 6845 \pm 412 \\ & (1635 \pm 98) \end{aligned}$ | $4.3 \pm 0.2$ | $\begin{aligned} & 24525 \pm 1596 \\ & (5858 \pm 381) \end{aligned}$ |  |
| Cyclists during the Tour de France, week 2 ( $n=4$ ) | M | $\begin{aligned} & 36025 \pm 1802 \\ & (8604 \pm 430) \end{aligned}$ | $\begin{aligned} & 6798 \pm 404 \\ & (1624 \pm 96) \end{aligned}$ | $5.3 \pm 0.6$ | $\begin{aligned} & 26275 \pm 854 \\ & (6276 \pm 204) \end{aligned}$ | [251] |
| cyclists during the Tour de France, week 3 ( $\mathrm{n}=4$ ) | M | $\begin{aligned} & 35650 \pm 2199 \\ & (8515 \pm 525) \end{aligned}$ | $\begin{aligned} & 6763 \pm 393 \\ & (1615 \pm 94) \end{aligned}$ | $5.3 \pm 0.3$ | $\begin{aligned} & 23225 \pm 1305 \\ & (5547 \pm 312) \end{aligned}$ |  |
| Highly trained endurance runners, 7 eumenorrceih and 2 oligomenorrheic ( $n=9$ ) | F | $\begin{aligned} & 12516 \pm 1737 \\ & (2989 \pm 415) \end{aligned}$ | NR | NR | $\begin{aligned} & 8527 \pm 1246 \\ & (2037 \pm 298) \end{aligned}$ | [252] |
| Elite distance runners ( $n=9$ ) | F | $\begin{aligned} & 11832 \pm 1306 \\ & (2826 \pm 312) \end{aligned}$ | $\begin{aligned} & 6025 \pm 950 \\ & (1439 \pm 227) \end{aligned}$ | $\begin{aligned} & 1.99 \pm \\ & 0.30 \end{aligned}$ | $\begin{aligned} & 9182 \pm 1951 \\ & (2193 \pm 466) \end{aligned}$ | [253] |
| Elite cross-country skiers during a pre-season period with highvolume training ( $n=4$ ) | F | $\begin{aligned} & 18300 \pm 2200 \\ & (4371 \pm 525) \end{aligned}$ | $\begin{aligned} & 5500 \pm 300^{*} \\ & (1314 \pm 72) \end{aligned}$ | $3.4 \pm 0.3$ | $\begin{aligned} & 18200 \pm 1900 \\ & (4347 \pm 454) \end{aligned}$ | [254] |
| Elite cross-country skiers during a pre-season period with highvolume training ( $n=4$ ) | M | $\begin{aligned} & 30200 \pm 4200 \\ & (7213 \pm 1003) \end{aligned}$ | $\begin{aligned} & 7600 \pm 300^{*} \\ & (1815 \pm 72) \end{aligned}$ | $4.0 \pm 0.5$ | $\begin{aligned} & 30200 \pm 4600 \\ & (7213 \pm 1099) \end{aligned}$ |  |
| Ultra-marathon running (7 month run around Australia) ( $n=1$ ) | M | $\begin{aligned} & 26088 \\ & (6231) \end{aligned}$ | $\begin{aligned} & \text { 6686* } \\ & \text { (1597) } \end{aligned}$ | 3.9 | NR | [255] |
| collegiate swimmers ( $n=8$ ) | $\begin{aligned} & \mathrm{M} \\ & / \\ & \mathrm{F} \end{aligned}$ | $\begin{aligned} & 14511 \pm 4153 \\ & (3466 \pm 992) \end{aligned}$ | NR | NR | $\begin{aligned} & 16308 \pm 2600 \\ & (3895 \pm 621) \end{aligned}$ | [256] |

Abbreviations: TEE, total energy expenditure; REE, resting energy expenditure (by indirect calorimetry); PAL, physical activity level (PAL = TEE/REE), EI, energy intake (self reported, weighed record, or food diary kept over); NR, not reported.
*REE estimated from equation

The analytical procedures involved in the DLW method are time-consuming, expensive, and involve complex methods and specialized technicians, excluding its routine use for EE assessment [137].

The use of energy intake from self reported measures has been suggested as an alternative to estimate energy requirements, since EI generally corresponds to DLW determine energy expenditure [257]. However, underreporting of nutritional intakes will result in difficult to accurately recommend energy requirements and consequently both health and performance may be affected. Conversely, the underestimation of EI is common in the athletic population which may be a concern to accurately estimate energy requirements [257]. From Table 2.7 it is possible to observe that EI is consistently underreported by athletes. In the beginning of the 1990's it was speculated that low EI in athletes with elevated EE could be a result of adaptation to chronically high levels of activity or due to genetic circumstances that became athletes as a 'more efficient machine' [258]. With the advent of the DLW method in the sports field it was possible to start deconstructing this theory and the cause to low EI was attributed to under-reporting [252]. This underreporting of energy expenditure has been the main focus of investigations regarding the energy balance in athletes [135, 247, 248, 252, 257]. In fact, exercise training itself has been shown to affect the accuracy of dietary recording in healthy non-obese adults and adolescents. Westerterp et al. [259] studied individuals at the beginning and end of a 40 -week training intervention programme. All subjects were previously non-exercisers and the initial difference between the subjects' self reported EI and EE from DLW was $25 \%$. However, by the end of the training program the discrepancy between the measurements increased to $219 \%$. Also, van Etten et al. [260] found an increased underreporting over the 18 -weeks of a weight-training program (from $21 \%$ to $34 \%$ ).

Objective methods to estimate energy requirements based on TEE from DLW have been developed and its accuracy in athletes has been analyzed [146, 261]. However these methods still present limitations in estimating athletes’ energy requirements. Koehler et al. [146] verified that the Sensewear Pro3 Armband (BodyMedia, Pittsburgh, PA), which is a portable electronic device that synchronically assesses biaxial accelerometry, body heat loss, and galvanic skin response did not provide valid results of TEE and AEE in endurance athletes due to an underestimation of EE at higher
exercise intensities. Nichols et al. [261] tested the accuracy of a combined heart rate and uniaxial motion sensor and observed that the equipment may have limited use estimating TEE, and therefore energy availability, in a sample of young female competitive runners. Silva et al. [247] verified that the Dietary Reference Intake method (based on an estimated physical activity level) though valid to assess energy expenditure in a group of basketball players, it was still inaccurate for determining individual energy requirements. In this framework, it is still necessary to validate new methods or to develop new algorithms for available physical activity electronic devices.

### 2.5.7. Body composition, Energy regulation and health in

## ATHLETES

The magnitude of body composition estimation on the athletic field goes far behind the impact on sports performance. The health status of the athlete is also a concern when investigating body composition in athletic populations.

Some physical activities are related to higher energy expenditure than others, and therefore energy requirements are distinct. A negative energy balance often occurs in weight sensitive sports (aesthetic sports, gravitational sports, or weight class sports). For example, female gymnasts, or ice dancers, for aestethic reasons, often have energy intakes as low as $4000 \mathrm{~kJ}(\sim 1000 \mathrm{kcal})$ to $8000 \mathrm{~kJ}(\sim 2000 \mathrm{kcal})$. In some situations, this intake is as low as only 1.2 to 1.4 times the REE, which is lower than sedentary people who, on average, expend 1.4 to 1.6 times the REE. However, this athletes may be involved in several hours of training per day, and therefore the energy expenditure is expected to be higher than sedentary people. Negative energy balance also occurs in upper limits of energy expenditure. Energy-related problems in endurance sports are completly different that the ones reported above. Well-trained endurance athletes can expend more than $4000 \mathrm{~kJ} / \mathrm{h}(\sim 1000 \mathrm{kcal} / \mathrm{h})$ for prolonged periods of time, resulting in highly daily energy expenditure. To mantain performance, energy stores must be replenished and energy balance must be restored, meaning that these athletes need to intake very large amounts of energy in periods of heavy training or competition. Athletes that are involved in sports like cycling, triathlon, cross-country sking, or
ultraendurance running are among the ones that are associated with a higher energy expenditure (table 2.7) [133].

Appart from the negative impact on sports performance, one of the main problems that are related to this negative energy balance is the possibility of health complications. In 1992, the concept of the female athlete triad was recognized when disordered eating, amenorrhea, and osteoporosis were verified in athletes from activities that emphasize a lean physique $[262,263]$. The female athlete triad is an interrelationship of menstrual dysfunction, low energy availability (with or without an eating disorder), and decreased bone mineral density (BMD), and it is relatively common among young women participating in sports [9, 147]. Weight loss in elite athletes is generally motivated by a desire to optimize performance, improving power to-weight ratio, making weight to compete in a certain weight category, or for aesthetic reasons in sports that emphasize leanness [41]. At these regard investigations have been conducted to understand the negative effects on bone metabolism and bone mass in sports where energy deficits may be extreme, and consequently bone demineralization may ensue [41].

In general, athletes tend to have a higher bone mineral density compared to nonathletes since physical activity has a beneficial effect on bone health [264]. However, regardless of similar weight bearing exercise, amenorrheic athletes present BMD than their eumenorrheic counterparts. In fact, amenorrheic athletes have $10 \%$ to $20 \%$ less lumbar spine BMD than eumenorrheic athletes [265-268]. Oligomenorrhea and amenorrhea can be detrimental to bone because they are hypoestrogenic states, and given that estrogens normally inhibits osteoclast activity, a lack of this hormone may cause disruption of bone remodelling and accelerated bone resorption [269]. Consequently, menstrual status in athletes may offset the beneficial effects of physical activity on bone health [147]. Amenorrhea can be caused by a variety of factors including energy deficiency [270]. In this respect the position stand of the American College of Sports Medicine [9] has recommended DXA assessments of BMD in athletes with history of hypoestrogenism, disordered eating or eating disorders for a cumulative total of 6 months or more, and/or a history of stress fractures or fractures from minimal trauma. In fact, investigations have been conducted in several weight sensitive sports to understand the adverse impact of a negative energy balance on bone health in weight
sensitive sports. These categories of weight sensitive sports include: aesthetic sports such as rhythmic and artistic gymnastics, figure skating, diving and synchronized swimming [271]; gravitational sports like long distance running, triathlon, ski jumping, high jumping or road cyclic [272, 273]; and weight class sports as wrestling, judo, boxing, taekwondo, jockeys, weight lifting and light-weight rowing [274-276].

### 2.6. The aim of the investigation

The present dissertation presents four research studies conducted under the scope of the body composition.

Study 1 (chapter 4) was conducted to solve a methodological problem regarding the assessment of body composition in individuals that are taller than the DXA scan area. This is particularly important in the athletic field, given that in some sports height is a major determinant of the athletic performance. In accordance, the objective of study 1 (chapter 4) was to validate an alternative procedure to assess body composition with DXA in participants larger than the scan area in athletic and non-athletic populations.

Given the importance of athletes meeting energy requirements both for health and sports performance it is emergent to provide accurate measurements of total energy expenditure in free living conditions. Therefore, the aim of the study 2 (chapter 5) was to validate an existing combined heart rate and motion sensor to estimate energy expenditure in a sample of basketball players at a pre-season training period.

Study 3 (chapter 6) was conducted to understand the changes that occur in body composition, at four-levels of analysis (i.e. molecular, cellular, tissue-system, and whole body) in the course of a season.

The last investigation, study 4 (chapter 7), was conducted to develop body composition percentiles at two levels, molecular and whole body composition levels, stratified by sex and sport.

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## Methodology

A brief description of the sample and study protocol will be provided in this chapter, however further specific details of the methods will be provided in each study (chapter 4 to 7).

### 3.1. Study design and sampling

All studies included in the present thesis were conducted within a project funded by the Portuguese Foundation for Science and Technology (grant: PTDC/DES/098963/2008), entitled Body Composition and Physical Performance Changes Over a Season in Elite Athletes. This project used an observational study with a follow-up over the season, including the assessment of body composition, energy expenditure, and physical tests at the beginning of a pre-season and close to the main national competition. While one study was conducted with this experimental design, for the remaining investigations a cross-sectional design was used. In Table 3.1 are summarized the basic characteristics of each study regarding sampling and design.

Table 3.1 Basic characteristics of each study: sampling and design

| Study | Sample | Sex | Age range | Design |
| :--- | :--- | :--- | :--- | :--- |
| $\mathbf{1}$ | athletes $(\mathrm{n}=31)$ | 13 M and 18 F | $16-29 \mathrm{yrs}$ | Cross-sectional |
|  | non-athletes $(\mathrm{n}=65)$ | 34 M and 31 F | $19-55 \mathrm{yrs}$ |  |
| $\mathbf{2}$ | Basketball players | 4 M and 8 F | $16-17 \mathrm{yrs}$ | Cross-sectional (pre-season) |
| $\mathbf{3}$ | Basketball players | 12 M and 11 F | $16-17 \mathrm{yrs}$ | Prospective (pre-season <br> competitive period) |
| $\mathbf{4}$ | Athletes from 21 sports | 264 F and 634 M | $16-50 \mathrm{yrs}$ | Cross-sectional (in-season) |

Abbreviations: F, female; M, males; yrs, years

### 3.2. Body composition measurements

### 3.2.1 ANTHROPOMETRY:

## Weight and height

Weight and height were assessed across all the studies presented in this dissertation (chapters 4 to 7). Participants were weighed without shoes to the nearest
0.01 kg minimal clothes on an electronic scale connected to the plethysmograph computer (BOD POD ${ }^{\oplus}$ COSMED, Rome, Italy). Based on 10 young active adults (5 males and 5 females), the coefficient of variation for body mass in our laboratory is $0.07 \%$. Height was measured to the nearest 0.1 cm with a stadiometer (Seca, Hamburg, Germany), according to standardized procedures [1]. Based on 9 male elite athletes the coefficient of variation for height is $0.04 \%$. Body mass index was calculated as weight $(\mathrm{kg})$ divided by the square of the height (m).

## Circumferences

In study 3 (chapter 6) and study 4 (chapter 7) circumferences were measured according to standardized procedures [1,2] with an anthropometric tape (Lufkin W606PM, Apex Tool Group, Sparks, Maryland U.S.A.) and reported to the nearest 0.1 cm . Circumferences measurements were conducted by two anthropometrists and the intra and inter coefficients of variation, calculated based on five highly active males, are presented in Table 3.2.

Table 3.2 Intra and inter coefficient of variations (CV) for circumferences measured by the two anthropometrists

| Circumferences | CV (measurer 1) | CV (measurer 2 ) | Inter measurers CV |
| :--- | :--- | :--- | :--- |
| Waist | $0.18 \%$ | $0.40 \%$ | 2.11 |
| Hip | $0.28 \%$ | $0.06 \%$ | 0.90 |
| Thigh | $0.15 \%$ | $0.27 \%$ | 1.02 |
| Calf | $0.19 \%$ | $0.09 \%$ | 0.67 |
| Arm | $0.22 \%$ | $1.49 \%$ | 1.31 |

Abbreviations: BMC, bone mineral content; BMD, bone mineral density; FM, fat-mass; FFM, fat-free mass; LST, lean soft tissue

Waist. Waist circumference was measured at minimal respiration by positioning an inelastic tape parallel to the floor and immediately above the iliac crest, according to the NIH procedures [2].

Hip. The subject stood straight with arms at the sides and feet together. The measurer squatted at the side of the subject so that the level of maximum extension of the buttocks could be seen. An inelastic tape was placed around the buttocks in a horizontal plane without compressing the skin [1].

Thigh (midthigh). With the subject standing, with the heels 10 cm apart and the weight evenly distributed between both feet, the measuring tape was placed horizontally around the thigh midway between the midpoint of the inguinal crease and the proximal border of the patella [1].

Calf. The subject stood with the feet about 20 cm apart and weight distributed equally on both feet. An inelastic tape measure was positioned horizontally around the calf and moved up and down to locate the maximum circumference in a plane perpendicular to the long axis of the calf. The level was marked so that the calf skinfold could be measured at the same level [1].

Arm. The measuring tape was placed in the midway between the lateral projection of the acromion process of the scapula and the inferior margin of the olecraneon process of the ulna. To locate the midpoint of the upper arm, the subject's elbow was flexed to $90^{\circ}$ with the palm facing superiorly and a small mark was made at the identified point [1].

Muscle circumferences. Arm, thigh, and calf circumferences muscle circumferences as circumference - (Л SKF) [3]. The circumferences were corrected for triceps, thigh, and calf SKF, respectively for arm, thigh and calf muscle circumferences.

## Skinfolds

Skinfold measurements were performed in study 3 (chapter 6) and study 4 (chapter 7) according to the procedures described by Lohman et al. [1]. The skinfold thickness was recorded to the nearest 0.1 cm and the sum of different combinations of skinfolds was used. Skinfolds measurements were conducted by two anthropometrists and the intra and inter coefficients of variation, calculated based on five highly active males, are presented in Table 3.3.

Subscapular. The subscapular skinfold was picked up on a diagonal, inclined infero-laterally approximately $45^{\circ}$ to the horizontal plane in the natural cleavage lines of the skin. The site is just below to the inferior angle of the scapula. The subject stood comfortably straight, with the upper extremities relaxed at the sides of the body. To locate the site, the measurer palpated the scapula, running the fingers inferiorly and laterally, along its vertebral border until the inferior angle was identified [1].

Table 3.3 Intra and inter coefficient of variations (CV) for skinfolds measured by the two anthropometrists

| Skinfolds | CV (measurer 1) | CV (measurer 2 ) | Inter measurers CV |
| :--- | :--- | :--- | :--- |
| Subscapular | $2.19 \%$ | $2.52 \%$ | $13.16 \%$ |
| Abdominal | $2.64 \%$ | $3.65 \%$ | $10.29 \%$ |
| Suprailiac | $1.96 \%$ | $3.59 \%$ | $11.08 \%$ |
| Thigh | $2.15 \%$ | $2.28 \%$ | $5.86 \%$ |
| Medial Calf | $5.66 \%$ | $4.53 \%$ | $3.15 \%$ |
| Triceps | $2.31 \%$ | $2.34 \%$ | $3.68 \%$ |
| Biceps | $0.00 \%$ | $0.00 \%$ | $10.54 \%$ |


#### Abstract

Abdominal. For the measurement of abdominal skinfold thickness, the subject relaxed the abdominal wall musculature as much as possible during the procedure and breathes normally. The subject stands straight with body weight evenly distributed on both feet. A site 3 cm lateral to the midpoint of the umbilicus and 1 cm inferior was selected and a horizontal skinfold was raised [1].

Suprailiac. The suprailiac skinfold was measured in the midaxillary line immediately superior to the iliac crest. The subject stood with feet together and in a straight position. The arms hanged by the sides, or, when necessary, they could be abducted slightly to improve access to the site. An oblique skinfold was grasped just posterior to the midaxillary line following the natural cleavage lines of the skin. It was aligned inferomedially at $45^{\circ}$ to the horizontal [1].


Thigh. The thigh skinfold site is located in the midline of the anterior aspect of the thigh, midway between the inguinal crease and the proximal border of the patella. The subject flexed the hip to assist location of the inguinal crease. The proximal reference point is on the inguinal crease at the midpoint of the long axis of the thigh. The distal reference point (proximal border of the patella) was located while the knee of the subject was extended. The thickness of a vertical fold was measured while the subject stood. The body weight was shifted to the other foot while the leg on the side of the measurement was relaxed with the knee slightly flexed and the foot flat on the floor [1].

Medial Calf. For the measurement of the medial calf skinfold the subject stood with the foot on a platform so that the knee and hip are flexed to about $90^{\circ}$. The level of the maximum calf circumference was marked on the medial aspect of the calf (see technique for calf circumference). The anthropometrist starts the measurement in front of the subject, raising a skinfold parallel to the long axis of the medial border of the calf, when viewed from the front, at a level slightly proximal to the marked site [1].

Triceps. The triceps skinfold was measured in the midline of the posterior aspect of the arm, over the triceps muscle, at a point midway between the lateral projection of the acromion process of the scapula and the inferior margin of the olecranon process of the ulna. The subject was measured standing with the arm hanging loosely and comfortably at the subject's side. A vertical fold was raised with the measurer standing behind the subject and placing the palm of the left hand on the subject's arm proximal to the marked level, with the thumb and index finger directed inferiorly. The site of measurement corresponded the midline posteriorly when the palm is directed anteriorly [1].

Biceps. Biceps skinfold was measured as the thickness of a vertical fold raised on the anterior aspect of the arm, over the belly of the biceps muscle. The skinfold was raised at the line marked for the measurement of triceps skinfold thickness and arm circumference. The subject stood, facing the measurer, with the upper extremity relaxed at the side, and the palm directed anteriorly. [1].

### 3.2.2. Hydration Status

The urine specific gravity (USG) was determined by a refractometer (Urisys 1100 Urine Analyzer, Roche, Portugal). The analyzer was calibrated with a control-Test (Chemstrip 10 MD ) every 7 days. After the dipsticks were inserted into the urine tubes they were placed and analyzed by the equipment, according to the manufacture standardized procedures. Based on test-retest in 10 young active adults the coefficient of variation for the USG technique in our laboratory is $0.1 \%$. This method was used in study 3 (chapter 6).

### 3.2.3. Total Body Water

Total body water was assessed by deuterium dilution technique using a stable Hydra gas isotope ratio mass spectrometer (PDZ, Europa Scientific, UK) in study 3 (chapter 7). After a 12 h fast, an initial urine sample was collected and immediately administrated a deuterium oxide solution dose $\left({ }^{2} \mathrm{H}_{2} \mathrm{O}\right)$ of 99.9 atom\% D (Sigma-Aldrich Chemistry) of $0.1 \mathrm{~g} / \mathrm{kg}$ of body weight, diluted in 50 mL of tap water. After a 4 h equilibration period, a new urine sample was collected. Abundances of ${ }^{2} \mathrm{H}_{2} \mathrm{O}$ in dilutions of the isotope doses were analyzed. Urine and diluted dose samples were prepared for analysis using the equilibration technique of Prosser and Scrimgeour [4]. The enrichments of equilibrated local water standards were calibrated against SMOW (Standard Mean Ocean Water). Based on delta SMOW, TBW was estimated including a $4 \%$ correction due to the recognized amount corresponding to deuterium dilution in other compartments [5]. The coefficient of variation based on test-retest using 10 participants was $0.3 \%$.

### 3.2.4. Extracellular Fluids

In study 3 (chapter 7) ECW was assessed by sodium bromide (NaBR) dilution. The subject was asked to drink $0.030 \mathrm{~g} / \mathrm{kg}$ of body weight of NaBr , diluted in 50 mL of deionized water. The NaBr concentration was measured by high-performance liquid chromatography (HPLC) (Dionex Corporation, Sunnyvale, CA) using a set of Ionpac AS9-HC Analytical column and Ionpac AG9-HC Guard column, and ASRS 300 suppressor. Baseline samples of plasma were collected before sodium bromide oral dose administration whereas enriched samples were collected 3 h post-dose administration.

The volume of ECW was calculated as:
$E C W(L)=\left[\right.$ dose $/\left(\right.$ post-fluid bromide $\left(\left[\mathrm{Br}^{-}\right]\right)-$pre-fluid $\left.\left(\left[\mathrm{Br}^{-}\right]\right)\right] \times 0.90 \times 0.95$
where 0.90 is a correction factor for intracellular bromide $(\mathrm{Br}-)$, found mainly in red blood cells, and 0.95 is the Donnan equilibrium factor [5].

Samples were pre-treated with acetonitrile to deproteinize and a correction for residual solid content in biological fluids was made ( 0.9745 and 0.996 for plasma and saliva, respectively).

Extracellular fluids were posterior calculated as ECW $\times(1 / 0.98)$. Based on testretest using 7 elite male athletes, the coefficient of variation for ECW was $0.4 \%$.

### 3.2.5. Intracellular Fluids

Intracellular fluids were calculated in study 3 (chapter 7) as the difference between TBW and ECW using the dilution techniques mentioned above (deuterium and sodium bromide, respectively).

### 3.2.6. Dual-Energy X-Ray Absorptiometry

Participants underwent a whole-body DXA scan according to the procedures recommended by the manufacturer on a Hologic Explorer-W, fan-beam densitometer (Hologic, Waltham, Massachusetts, USA). The equipment measures the attenuation of X-rays pulsed between 70 and 140 kV synchronously with the line frequency for each pixel of the scanned image. Following the protocol for DXA described by the manufacturer, a step phantom with six fields of acrylic and aluminium of varying thickness and known absorptive properties was scanned to serve as an external standard for the analysis of different tissue components. The same technician positioned the participants, performed the scan and executed the analysis (software QDR for Windows version 12.4, Hologic, Waltham, Massachusetts, USA) according to the operator's manual using the standard analysis protocol. The DXA measurements included whole body or regional measurements of bone mineral content (studies 1,3 , and 4 ), bone mineral density (study 4), absolute fat mass (studies 1, 2, and 4), percent FM (studies 1, 2, and 4), fat-free mass (studies 2 and 4), and lean soft tissue (studies 1,3 , and 4).

The coefficients of variation in our laboratory based on 10 young active adults (five males and five females) are presented in Table 3.4.

Table 3.4 Coefficients of variation in our laboratory for Dual-energy X-ray Absorptiometry measurements

|  | Whole-body | Sub-total | Appendicular | Trunk |
| :--- | :--- | :--- | :--- | :--- |
| BMC | $1.3 \%$ | $0.9 \%$ | $0.9 \%$ | $2.5 \%$ |
| BMD | $1.4 \%$ |  |  |  |
| Absolute FM | $1.7 \%$ | $1.8 \%$ | $2.8 \%$ | $4.3 \%$ |
| Percent FM | $1.6 \%$ | $1.7 \%$ | $2.1 \%$ | $3.6 \%$ |
| FFM | $0.8 \%$ | $0.6 \%$ | $1.6 \%$ | $1.2 \%$ |
| LST | $0.8 \%$ | $0.6 \%$ | $1.2 \%$ | $1.3 \%$ |

Abbreviations: BMC, bone mineral content; BMD, bone mineral density; FM, fat-mass; FFM, fat-free mass; LST, lean soft tissue

In the study 3 (chapter 6) skeletal muscle (SM) mass was estimated for the tissue-system level as [6]:
$S M(k g)=[1.19 \times \operatorname{ALST}(k g)]-1.65$

Where ALST is appendicular lean soft tissue assed by dual-energy X-ray absorptiometry.

### 3.2.7. BODY CELL MASS

At the cellular level, body cell mass was estimated in study 3 (chapter 6) according to Shen et al. [7] as:
$B C M(k g)=L S T_{D X A}-(E C F+E C S)$

Where $\mathrm{LST}_{\mathrm{DXA}}$ is lean soft tissue from DXA ( kg ), ECF is extracellular fluids obtained by the dilution technique ( kg ), and ECS is extracellular solids calculated as $1.732 \times \mathrm{Mo}(\mathrm{kg})[\mathrm{Mo}(\mathrm{kg})=\mathrm{BMC}(\mathrm{kg}) \times 1.0436]$.

The propagation measurement error associated with measurement of BCM was estimated by assuming an average body composition and measurement precision of each method [8]. The calculations are described in detail in chapter 6 and the precision is 0.5 kg for BCM .

### 3.2.8. Body Volume

Measures of body volume were conducted in study 3 (chapter 6) to use a 4component model. Body volume (BV) was assessed by air displacement
plethysmography (BOD POD ${ }^{\odot}$ COSMED, Rome, Italy). After voiding their bladder, each subject was weighed to the nearest gram while wearing a swimsuit. The ADP device was calibrated according to the manufacturer's instructions. The effects of clothing and hair were accounted for by using a bathing suit and a swim cap. Finally, thoracic gas volume ( $\mathrm{T}_{\mathrm{GV}}$ ) was measured in the $\mathrm{BOD} \mathrm{POD}^{\circledR}$ by using a technique common to standard pulmonary plethysmography called the "panting maneuver." While wearing a nose clip, the subjects breathed through a tube; after 2 to 3 normal breaths, the airway occluded for 3 seconds at mid-exhalation. During this time, the subject was instructed to gently puff against the occlusion by alternately contracting and relaxing the diaphragm. All measurements were conducted with software version 1.68. The coefficient of variation for body volume, based on test-retest using 10 young active adults ( 5 males and 5 females), were $0.4 \%$ and 0.20 L , respectively.

### 3.2.9. Four-component Model.

A four-component model was used in study 3 (chapter 6) to assess the molecular level of body composition, calculated after using the total-body soft tissue mineral (Ms) component obtained as $\mathrm{Ms}=0.0129 \times$ TBW [9]. The model is described as follows:
$F M(k g)=2.748 \times B V-0.699 \times T B W+1.129 \times M o-2.051 \times B M$

Where BV is body volume (L), TBW is total body water $(\mathrm{kg})$, Mo is bone mineral $(\mathrm{kg})[\mathrm{Mo}(\mathrm{kg})=\mathrm{BMC}(\mathrm{kg}) \times 1.0436]$, and $B M$ is body mass ( kg ).

The FFM was then calculated as BM minus FM.
The propagation measurement error associated with measurement of FM from the 4 -component model was estimated by assuming an average body composition and measurement precision of each method [8]. The calculations are described in detail in chapter 6 and the precision is 0.7 kg for FM .

## Calculation of Density of Fat-free Mass

The $\mathrm{FFM}_{\mathrm{D}}$ was estimated from TBW, Mo, Ms and protein (protein is equal to BM minus FM from the 4C model, TBW, Mo and Ms), contents of FFM (estimated as

BM minus FM from the 4C model) and their densities ( $0.9937,2.982,3.317$, and 1.34 $\mathrm{g} / \mathrm{cm}^{3}$ ), for TBW, Mo, Ms and protein, respectively,
$\mathrm{FFM}_{\mathrm{D}}=1 /\left[\left(\mathrm{TBW} / \mathrm{TBW}_{\mathrm{D}}\right)+\left(\mathrm{Mo} / \mathrm{Mo}_{\mathrm{D}}\right)+\left(\mathrm{Ms} / \mathrm{MS}_{\mathrm{D}}\right)+\left(\right.\right.$ protein protein $\left.\left._{\mathrm{D}}\right)\right]$
Where ${ }_{D}$ is density, FFM is fat-free mass, TBW is total body water, Mo is bone mineral, and Ms is total-body soft tissue mineral.

### 3.3. Energy expenditure measurements

Energy expenditure measurements were conducted in study 2 (chapter 5). Participants came to the laboratory in the morning having for at least 12-hours, refraining from vigorous exercise for at least 14 -hours and did not consume caffeine, alcohol or stimulant beverages for at least 24-hours before the testing begin at 8:00 a.m.

### 3.3.1. TOTAL ENERGY EXPENDITURE WITH DOUBLY LABELLED WATER.

Doubly labelled water (DLW) was administered in the morning of the body composition assessment, both at the pre-season and at the competitive training period. The TEE was measured by an established procedure [10]. Briefly, the subjects were weighed in the morning and baseline urine was collected. At the pre-season an oral dose of $0.8 \mathrm{~g} / \mathrm{kg}$ of TBW of a 10 atom $\% \mathrm{H}_{2}{ }^{18} \mathrm{O}$ (Taiyo Nippon Sanso Corporation, Tokyo, Japan) (assuming TBW is $61 \%$ of body mass), and $0.16 \mathrm{~g} / \mathrm{kg}$ of TBW of 99.9 atom $\%$ ${ }^{2} \mathrm{H}_{2} \mathrm{O}$ (Sigma Aldrich, Co, St Louis, MO, USA), diluted in 50 ml of tap water was administered to the subjects at $7.00 \mathrm{a} . \mathrm{m}$. During the morning, post-dose urine samples were collected. Urine samples included the collection of day 0 at baseline, 4 and 5 h post-dose, and at day 7 , the first urine in the morning and 1 h , and frozen ate $-20^{\circ}$ for posterior analyses. These urine samples were prepared and filled with the equilibration gas.

Equilibration period was 3 days and 8 h , for ${ }^{2} \mathrm{H}$ and ${ }^{18} \mathrm{O}$, respectively. Samples were analyzed in duplicates and calibrated against standard mean ocean water (SMOW), using Hydra isotope ratio mass spectrometer (PDZ, Europa Scientific, UK). Energy expenditure by the DLW method was calculated from a modified Weir's equation by use from DLW and calculated from the food quotient obtained by dietary intake records [10]. The coefficient of variation for total energy expenditure, based on test- retest
using 10 subjects is $4.3 \%$. Activity energy expenditure was calculated as TEE-RMR$0.1 \times$ TEE (assuming the thermic effect of food is $\sim 10 \%$ of TEE) and physical activity level (PAL) was determined as TEE/REE.

### 3.3.2. Total energy expenditure with combined heart rate and

## MOTION SENSOR.

Energy expenditure simultaneously evaluated with combined HR and motion sensor (Actiheart, Cambridge Neurotechnology Ltd, Cambridge, UK). The monitor was worn using ECG pads in the chest during the same 7-day period that the DLW assessment took place. Participants performed an 8 -min step-test (height: 215 mm ), the stepping speed ramps linearly increased from 15-33 step cycles/min, providing individual HR-EE relationship calibration. From the individual step-test calibration estimated $\mathrm{VO}_{2} \max$ was derived by the software. The device was started with $60-\mathrm{s}$ epochs and participants were asked to wear the monitor at all times (even during sleep hours) for the 7consecutive days the DLW assessment were taking place. Data from the monitors were downloaded into to the commercial software (v.4.0.46). The software algorithm allowed data cleaning, recovering, and interpolation of missing and noisy HR. Only participants with 3 -valid days were considered for data analysis. A valid day was considered when we had at least $70 \%$ of the day ( 1008 min ) with records and not more than $10 \%$ of the registered timed with HR recovered by the software. Moreover, if the participants had invalid data during the training hours (registered in a diary) the day was not considered valid.AEE was estimated using energy models, available in the commercial software:
$\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ : individual HR calibration model (Group CalJAP2007/Step HR[11]), with HR and accelerometry data;

ACC+HR group: group HR calibration model(Group CalJAP2007[11]) with HR and accelerometry data;
$\mathrm{HR}_{\text {flex }}$ : individual HR calibration model (Group CalJAP2007/Step HR[11]), with HR data;

ACC: accelerometry data [11].

TEE was estimated adding to the estimated AEE, the thermic effect of food ( $\sim 10 \%$ of TEE) and REE using the Schofield equation[12], as suggested by the manufacturer.

### 3.3.3. RESTING ENERGY EXPENDITURE (REE)

Measurements were performed between 8:00 and 10:00 a.m., and regarding the female athletes, on one of the days within middle-to-late follicular phase of menstrual cycle ( $<10$ days since last menstruation). Prior to the REE measurements, the subjects lied supine for 10 min covered with a blanket in a quiet room at an environmental temperature and humidity of $\pm 22^{\circ} \mathrm{C}$ and $40-50 \%$, respectively. The REE was determined by an open-circuit indirect calorimetry through a portable gas analyzer (K4b ${ }^{2}$, Cosmed, Rome, Italy). Expired gases were analyzed continuously, breath by breath. After the mask was placed in the subject face, oxygen consumption $\left(\dot{V} O_{2}\right)$ and $\mathrm{CO}_{2}$ production $\left(\dot{V} \mathrm{CO}_{2}\right)$ were measured for an additional 20-min period. Outputs of $\dot{V} O_{2}, \dot{V} C O_{2}$, respiratory exchange ratio (RQ), and ventilation were collected and averaged over 1-min intervals for data analysis. The first and the last 5 -min of data collection were discarded and the mean of a 5-min steady state interval between the $5^{\text {th }}$ and the $25^{\text {th }}$ min with RQ between 0.7 and 1.0 was used to calculate REE. Steady state was defined as a 5 -min period with $\leq 10 \% \mathrm{CV}$ for $\dot{V} O_{2}$ and $\dot{V} C O_{2}$ [13]. The mean $\dot{V} O_{2}$ and $\dot{V} C O_{2}$ of $5-\mathrm{min}$ steady states were used in Weir equation [14] and the period with the lowest REE was considered. Before each test, the $\mathrm{O}_{2}$ and $\mathrm{CO}_{2}$ analyzers were calibrated using standard calibration gases of known concentration ( $16.7 \% \mathrm{O}_{2}$ and $5.7 \%$ $\mathrm{CO}_{2}$ ). The calibration of the turbine flowmeter of the $\mathrm{K} 4 \mathrm{~b}^{2}$ was performed using a 3-L syringe (Quinton Instruments, Seattle, WA, USA) according to the manufacturer's instructions. The coefficient of variation for REE, based on test-retest using five subjects, is approximately $10 \%$.

### 3.4. Performance measurements

### 3.4.1 HANDGRIP.

Maximal isometric forearm strength (HGrip) was determined using a handgrip dynamometer (Jamar, Sammons Preston, Inc, Bolingbrook, IL, U.S.A.) with visual feedback. The dynamometer was adjusted to each subject's dominant hand with each trial lasting approximately 5 -seconds. The best of three maximal trials was used for data analysis. The same adjustment of the dynamometer was used for all tests for each subject. The handgrip test was performed in study 3 (chapter 6).

### 3.4.2. Vertical Jump.

Explosive power of the lower limbs was assessed by performing a countermovement jump abalakov (CMJ) in a custom contact platform (BioPlux System, version 1.0, Lisbon, Portugal). Participants were given detailed instructions and performed 2 trial jumps ( $\sim 50 \%$ of maximal height) with a resting period of 15 -seconds in between. The starting position was from upright standing position. They were then instructed to flex their knees $\left(90^{\circ}\right)$ as quickly as possible and then jump as high as possible with arm swing in the ensuing concentric phase. Subjects performed 3 jumps, with a 30 -seconds resting period and the jump with the greatest high was selected. The vertical jump test was performed in study 3 (chapter 6).

### 3.4.3. MAXIMAL OXYGEN CONSUMPTION.

Maximal oxygen consumption $\left(\mathrm{VO}_{2 \text { max }}\right)$ measurement was performed with a continuous, progressive treadmill running protocol in a laboratory $\left(21-22^{\circ} \mathrm{C}\right.$, relative humidity of $50 \%$ ). Following a 2 -min warm-up (males: $0 \%$ grade; $7 \mathrm{~km} / \mathrm{h}$ speed; females: $0 \%$ grade; $6 \mathrm{~km} / \mathrm{h}$ speed), subjects ran for 2-min (males: $0 \%$ grade; $9 \mathrm{~km} / \mathrm{h}$ speed; males: $0 \%$ grade; $8 \mathrm{~km} / \mathrm{h}$ speed). Speed and grade were incremented $1 \mathrm{~km} / \mathrm{h}$ and $1 \%$, respectively, every $2-\mathrm{min}$, until exhaustion. Subjects received verbal encouragement and where instructed to exercise to volitional fatigue. Breath-by-breath gases were continuously analyzed with an open-circuit spirometry system (Quark b², Cosmed, Rome, Italy). Heart rate was continuously measured during the test (Polar

Electro Oy, Kempele, Finland). $\mathrm{VO}_{2 \max }$ was attained when at least two of the following three criteria were achieved: no increase in $\mathrm{VO}_{2 \max }$ despite further increases in work rate, a heart rate at or above age predicted maximum, and/or a RER $\geq 1.0$. Maximal oxygen consumption was measured in study 2 (chapter 5).

### 3.5. Statistical analysis

Data analysis was performed using the following softwares: IBM SPSS Statistics (SPSS Inc., an IBM Company, Chicago, Illinois, USA) version 19.0 or 21.0 (studies 1, 2, 3 and 4); MedCalc Statistical Software (Mariakerke, Belgium) version 11.1.1.0 (studies 1 and 2); and R version 2.14.2 [15] (study 4).

The statistical procedures common to all studies are presented in this section (Chapter 4 to 7), as follows:

Descriptive statistics including means and standard deviation were performed for all outcome measurements. Normality of the outcome variables was analyzed using the Kolmogorov-Smirnov test or the Shapiro Wilk-test. Mean comparisons for two groups were performed using independent sample T-test or the alternative MannWhitney tests while comparisons for three or more groups was performed using Oneway ANOVA or the alternative Kruskall-Wallis test. Paired sample t-tests, or the alternative non-parametric Wilcoxon-test were used to compare measures from paired samples.

Additionally we included statistical analyses that were specific to each of the studies, according to the objectives that were proposed for each investigation.

In study 1 (chapter 4) and study 2 (chapter 5) specific statistical procedures were used to test the accuracy of the alternative methods as described in detail in each chapter.

In study 3 (chapter 6) we additionally included one sample $t$-tests to test changes that significantly differed from zero and to compare group means with the reference values based on cadaver analysis. Also, Pearson correlations were used to analyse de association between body composition parameters and between body components with performance variables.

In study 4 (chapter 7) analyses were performed to complete different tasks: 1) to estimate the reference percentiles for each outcome, stratified by sex and sport; 2) to test whether or not the mean for each outcome differs by sex, stratified by sport; 3), to identify sports within each outcome for which the mean value is different from the others (if any), stratified by sex; and 4) to test for an association between anthropometric variables and DXA outcomes. The specific procedures to complete each task are described in detail in study 4 (chapter 7).

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# Body composition in taller individuals using DXA: a validation study for athletic and non-athletic <br> populations ${ }^{1}$ 

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# Body composition in taller individuals using DXA: A validation study for athletic and non-athletic populations 

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### 4.1. Abstract

Aim. Dual energy X-ray absorptiometry (DXA) cannot be used to evaluate participants taller than the scan area. We aimed to analyse the accuracy of bone mineral content, fat mass, and fat-free mass assessed with DXA whole-body scan and from the sum of two scans (head and trunk plus limbs).

Methods. Participants were 31 athletes ( 13 males and 18 females) and 65 nonathletes ( 34 males and 31 females), that fit within the DXA scan area. Three scans were performed using a Hologic Explorer-W fan-beam densitometer: a whole-body scan used as the reference; a head scan; and a trunk and limbs scan. The sum of the head scan and the trunk and limbs scan was used as the alternative procedure. Multiple regression and agreement analysis were performed.

Results. Non-significant differences between methods were observed for fat mass ( 0.06 kg ) and lean soft tissue ( 70.07 kg ) while bone mineral content from the alternative procedure differed from the reference scan ( 0.009 kg ). The alternative procedure explained $>99 \%$ of the variance in the reference scan and low limits of agreement were observed. Precision analysis indicated low pure errors and the higher coefficients of variation were found for fat mass (whole-body: $3.70 \%$; subtotal: $4.05 \%$ ).

Conclusions. The method proposed is a valid and simple solution to be used in individuals taller than the DXA scan area, including athletes engaged in sports recognised for including very tall competitors.

Keywords: fat mass, bone mineral content, lean soft tissue, athletes, dual-energy X-ray absorptiometry

### 4.2. Introduction

Dual-energy X-ray absorptiometry (DXA) is a widely accepted method to assess bone mineral content, fat mass, and lean soft tissue [1-4], either in clinical, research or in athletic settings.

Particularly within the athletic field, body composition assessment may help to optimise competitive performance and assess the effects of training [5]. Therefore, accurate body composition measurements are of considerable interest to athletes and coaches [5, 6].

The past decades in the history of DXA have been characterised by technological advances that allowed for a time-efficient and minimal-risk method of assessing whole-body and regional body composition [7]. Despite DXA's accuracy, precision, reliability, high speed, and non-invasiveness [7-9], one of its main limitations is the fact that a whole-body scan can only be performed in individuals shorter than the scan area, which varies between 185 and 197 cm , depending on the equipment [10]. This limitation particularly affects athletes involved in sports where height is a major factor of performance, such as basketball and volleyball.

Few studies have proposed alternatives to body composition assessment in individuals taller than the DXA scan area [10, 11]. Silva et al. [11] used correction models for bone mineral content, fat mass, and lean soft tissue, and indicated that a single scan with the knees bent can be performed for a specific DXA instrument (Hologic QDR-1500). Regardless of the added-value of this reported study a pencilbeam mode was used with the knees bent at an angle of 908. This is not a useful approach since for some scanners the distance between the scanning arm and the examination table may be lower, not allowing the participants to bend their knees. Also using Hologic equipment (pencilbeam mode, Hologic 1000) another study proposed two summing methods of partial scans, separating one at the neck and one at the hip to estimate the whole-body scan [10]. They observed that, although both methods were valid, the technique where the neck was set to divide the body scan in two parts provided more accurate estimates of bone and soft tissue. Nevertheless this validation study only included 19 non-athlete participants.

The accuracy and usefulness of an easier methodology to determine body composition in individuals taller than the DXA scan area, using a large and diverse sample of athletes and non-athletes, is of higher interest and applicability. The aim of this study was to analyse the accuracy of DXA in assessing bone mineral content, fat mass, and lean soft tissue with the sum of two scans (head and trunk plus limbs), using a whole-body scan as the reference criteria, in a sample of male and female athletes and non-athletes.

### 4.3. Methods

### 4.3.1. PARTICIPANTS

Body composition was measured in 31 athletes ( 13 males and 18 females) and 65 non-athletes ( 34 males and 31 females), who volunteered to participate in this study. All the participants included in this study were healthy, non-obese (categorised as a body mass index $<30 \mathrm{~kg} / \mathrm{m}^{2}$ ) and fit within the DXA scan area ( $<195 \mathrm{~cm}$ ). Participants ranged in age from 16 to 55 years old, height from 152.8 to 186.8 cm , body mass from 41.9 to 98.6 kg , body mass index from 17.0 to $29.7 \mathrm{~kg} / \mathrm{m}^{2}$, and percent fat mass from 6.9 to $35.9 \%$.

The athletic group was comprised of national elite athletes of different sports: triathlon, judo, rowing, track and field athletics, pentathlon, tennis, basketball, and wrestling.

Participants were informed about the possible risks of the investigation before giving their written informed consent to participate. All procedures were approved by the Ethics Committee of the Faculty of Human Kinetics, Technical University of Lisbon, and conducted in accordance with the declaration of Helsinki for human studies of the World Medical Association [12].

### 4.3.2. Body Composition Measurements

After a 3-h fast, participants came to the laboratory where all measurement procedures were carried out. In brief, the procedures are described as follows:

## Anthropometric measurements

Participants were weighed to the nearest 0.01 kg wearing minimal clothes on an electronic scale connected to the plethysmograph computer (BODPOD ${ }^{\oplus}$, COSMED, Rome, Italy). Height was measured to the nearest 0.1 cm with a stadiometer (Seca, Hamburg, Germany), according to the standardised procedures described elsewhere [13].

## Dual energy X-ray absorptiometry

To assess bone mineral content, fat mass, and lean soft tissue, DXA measurements were performed with a total body scan Hologic Explorer-W, fan-beam densitometer, software QDR for Windows version 12.4 (Hologic, Waltham, Massachusetts, USA). The equipment measures the attenuation of X-rays pulsed between 70 and 140 kV synchronously with the line frequency for each pixel of the scanned image. Following the protocol for DXA described by the manufacturer, a step phantom with six fields of acrylic and aluminium of varying thickness and known absorptive properties was scanned to serve as an external standard for the analysis of different tissue components. Following the protocol described by the manufacturer, we performed a whole-body scan used as the reference and two additional scans in order to attend the purpose of this study (Figure 4.1), specifically: a) a head scan, where the DXA scan length (approximately 80 cm ) was set at a height sufficient to scan from the top of the head to the lower jaw; and b) a trunk and limbs scan, where the participant was positioned with the head slightly out of the scan area. The scan length was set as the normal length for the whole-body scan ( 195 cm ) and for the trunk and limbs scan. The sum of head and trunk plus limbs was used as an alternative procedure to assess bone mineral content, fat mass and lean soft tissue. For data analysis whole-body (limbs, trunk, and head) and subtotal (limbs and trunk) measurements were considered.

The same technician positioned the participants, performed the three scans and executed the analysis according to the operator's manual using the standard analysis protocol. The technician also set the delimitation to include and exclude the head in the head scan and the trunk and limbs scan, respectively (Figure 4.1). Based on 10 young active adults (five males and five females), the coefficient of variation in our laboratory for bone mineral content is $1.3 \%$, for fat mass $1.7 \%$, and for lean soft tissue $0.8 \%$. The
technical errors of measurement are 0.03 kg for bone mineral content, 0.21 kg for fat mass, and 0.34 kg for lean soft tissue.


Figure 4.1. Participants' position and delimitation marks in DXA scan area, for the reference (a), head (b) and trunk and limbs (c) scans.

### 4.3.3. STATISTICAL ANALYSIS

Data analysis was performed using IBM SPSS Statistics version 19.0, 2010 (SPSS Inc., an IBM

Company, Chicago, Illinois, USA) and the MedCalc Statistical Software (MedCalc Software, Mariakerke, Belgium). Descriptive statistics including means $\pm$ standard deviation were performed for all the measurements. Normality was tested using the Kolmogorov-Smirnov test. Independent sample t-tests or the alternative Mann-Whitney tests were used for sex and athletic status comparisons. Paired sample ttests, or the alternative non-parametric Wilcoxon-test were used to compare bone mineral content, fat mass, and lean soft tissue values from the alternative procedure with
the reference scan. In order to test the accuracy of the body components assessed by the alternative scan, multiple regression analyses were performed. The interaction terms between sex by each main predictor (bone mineral content, fat mass, and lean soft tissue from the alternative procedure) and athletic status by the aforementioned predictors were tested in separate models for bone mineral content, fat mass, and lean soft tissue assessed by the reference scan (dependent variables). If non-significant interaction terms were found further analysis would be conducted using the whole sample. Linear regression models, separately, for whole-body and subtotal bone mineral content, fat mass, and lean soft tissue using the reference scan as the dependent variables and bone mineral content, fat mass, and lean soft tissue estimated by the alternative procedure, respectively, as the independent variables were performed. Normality, homogeneity, and homoscedasticity of the residuals were analysed.

The concordance correlation coefficient was analysed to evaluate the degree to which pairs of
observations fall on the $45^{\circ}$ line through the origin [14]. The concordance correlation coefficient ( $\rho \mathrm{c}$ ) contains a measurement of precision $\rho$ and accuracy ( $\rho \mathrm{c}=$ $\rho \mathrm{Cb}$ ) where $\rho$ is the Pearson correlation coefficient, which measures how far each observation deviates from the best-fit line, and Cb is a bias correction factor that measures how far the best fit line deviates from the $45^{\circ}$ line through the origin, and is a measure of accuracy. The differences between the methods (bias) and the $95 \%$ limits of agreement were used to analyse the agreement between the methods. The pure error and the coefficient of variation were used as measures of precision [15].

Stepwise linear regression analyses were performed to understand the potential covariates that could improve the explanation of the variability of the reference scan, when using the alternative procedure. The tested variables were age, sex, athletic status, and interactions.

Statistical significance was set at $\mathrm{p}<0.05$ (2-tailed) for all analyses.

### 4.4. Results

Participants' characteristics are described in Table 4.1.
Table 4.1. Descriptive characteristics (mean $\pm$ SD) of athletes, non-athletes, and whole sample

|  | Athletes |  |  | Non-Athletes |  |  | Whole sample |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Variables | Male $(n=13)$ | $\begin{aligned} & \text { Female } \\ & (n=18) \end{aligned}$ | Total $(n=31)$ | Male $(n=34)$ | $\begin{aligned} & \text { Female } \\ & (\mathrm{n}=31) \end{aligned}$ | Total $(n=65)$ | Male $(n=47)$ | $\begin{aligned} & \text { Female } \\ & (n=49) \end{aligned}$ | $\begin{aligned} & \text { Total } \\ & \text { ( } \mathrm{n}=96 \text { ) } \end{aligned}$ |
| Age (yrs) | $19.5 \pm 3.5$ | $19.7 \pm 4.0$ | $19.6 \pm 3.7^{*}$ | $25.7 \pm 7.5$ | $28.0 \pm 9.1$ | $26.8 \pm 8.3^{*}$ | $24.0 \pm 7.1$ | $24.9 \pm 8.6$ | $24.5 \pm 7.9$ |
| BM (kg) | $72.0 \pm 6.8$ | $63.3 \pm 7.3$ | $67.0 \pm 8.2$ | $73.2 \pm 9.1$ | $58.8 \pm 9.7$ | $66.3 \pm 11.8$ | $72.8 \pm 8.5^{5}$ | $60.4 \pm 9.1$ | $66.5 \pm 10.7$ |
| Height (cm) | $180.1 \pm 5.3$ | $171.6 \pm 7.3$ | $175.2 \pm 7.7^{*}$ | $177.0 \pm 9.1$ | $163.5 \pm 6.1$ | $170.6 \pm 9.2^{*}$ | $177.8 \pm 6.3^{5}$ | $166.5 \pm 7.6$ | $172.1 \pm 9.0$ |
| BMI ( $\mathrm{kg} / \mathrm{m}^{2}$ ) | $22.1 \pm 1.1$ | $21.5 \pm 2.1$ | $21.8 \pm 1.8$ | $23.4 \pm 2.6$ | $21.9 \pm 2.8$ | $22.7 \pm 2.8$ | $23.0 \pm 2.3^{5}$ | $21.8 \pm 2.6$ | $22.4 \pm 2.5$ |
| FM (\%) | $12.7 \pm 3.2$ | $21.1 \pm 3.4$ | $17.6 \pm 5.4^{*}$ | $16.7 \pm 4.6$ | $27.5 \pm 6.0$ | $21.8 \pm 7.6^{*}$ | $15.6 \pm 4.6^{5}$ | $25.2 \pm 6.0$ | $20.5 \pm 7.2$ |
| FM (kg) | $9.0 \pm 2.8$ | $13.3 \pm 3.2$ | $11.5 \pm 3.7^{*}$ | $12.2 \pm 4.4$ | $16.2 \pm 5.4$ | $14.1 \pm 5.3^{*}$ | $11.3 \pm 4.2^{5}$ | $15.2 \pm 4.9$ | $13.3 \pm 4.9$ |
| LST (kg) | $58.7 \pm 4.3$ | $46.7 \pm 4.6$ | $51.7 \pm 7.5$ | $57.0 \pm 6.5$ | $39.6 \pm 5.1$ | $48.7 \pm 10.5$ | $57.5 \pm 6.0^{5}$ | $42.2 \pm 6.0$ | $49.7 \pm 9.7$ |
| BMC (kg) | $2.59 \pm 0.30$ | $2.45 \pm 0.39$ | $2.51 \pm 0.35$ | $2.75 \pm 0.35$ | $2.07 \pm 0.33$ | $2.42 \pm 0.48$ | $2.70 \pm 0.34^{5}$ | $2.21 \pm 0.39$ | $2.45 \pm 0.44$ | $\overline{\text { Abbreviations: } B M \text {, body mass; BMI, body mass index; FM, fat mass; LST, lean soft tissue; BMC, bone mineral content. }}$ * Significant differences between athletes and non-athletes ( $\mathrm{p}<0.05$ ).

${ }^{5}$ Significant differences between sexes ( $\mathrm{p}<0.05$ ).

Since no interactions were observed between each main independent predictor (bone mineral content, fat mass, and lean soft tissue from the alternative procedure) with sex ( $p=0.40, p=0.12$ and $p=0.24$, respectively) and athletic status $(p=0.10, p=$ 0.83 and $p=0.80$, respectively) obtained in separate models (bone mineral content, fat mass, and lean soft tissue from the reference scan), the entire sample was used to analyse the accuracy of the alternative method.

Small but significant differences between the reference and the alternative scans were only observed for whole-body ( 0.009 kg ) and subtotal bone mineral content $(0.008$ kg ). Non-significant differences between the reference and the alternative scans, both for whole-body (fat mass: 0.06 kg , lean soft tissue: -0.07 kg ) and subtotal DXA's results (fat mass: 0.06 kg , lean soft tissue: -0.08 kg ), were observed (Table 4.2).

Figure 4.2 (panel A) represents the associations between the reference (dependent variable) and the alternative (independent variable) scans for whole-body bone mineral content, fat mass, and lean soft tissue. Linear regression analysis showed that the three components, measured by the alternative procedure, explained $>99 \%$ of the variance of the whole-body (Figure 4.2) and subtotal bone mineral content, fat mass, and lean soft tissue, assessed by the reference scan (Table 4.2). Models presented a low standard error of estimation both for whole-body and subtotal bone mineral content, fat mass, and lean soft tissue. The concordance correlation coefficient values were 0.997, 0.995 , and 0.998 correspondingly for bone mineral content, fat mass, and lean soft tissue, as indicated in Table 4.2.

Considering the precision (Table 4.2) of the alternative procedure, low pure errors were found both for whole-body (bone mineral content: 0.034 kg ; fat mass: 0.497 kg , and lean soft tissue: 0.536 kg ) and subtotal results (bone mineral content: 0.031 kg ; fat mass: 0.496 kg , and lean soft tissue: 0.550 kg ). The higher coefficient of variation was observed for fat mass (whole-body: $3.70 \%$ and subtotal: $4.05 \%$ ), while the lower values were verified for lean soft tissue (whole-body: $1.09 \%$ and subtotal: $1.20 \%$ ).
Table 4.2. Validation of whole-body and subtotal bone mineral content, fat mass, and fat-free mass ( $n=96$ )

|  | Whole-body |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- |
|  | BMC |

Abbreviations: BMC, bone mineral content; FM, fat mass; LST, lean soft tissue; ${ }^{2}$, coefficient of determination; SEE, standard error of estimation; CCC, concordance correlation coefficient; Bias, differences between the methods; LoA, limits of agreement; PE, pure error; CV, coefficient of variation.
*Significantly different from the reference scan.


Figure 4.2. Linear regression (left panel) for whole-body bone mineral content, fat mass, and lean soft tissue estimation using the reference method and bone mineral content, fat mass, and lean soft tissue using the sum of head plus trunk and limbs scan (Panel A) and the respective residual plots (Panel B).

We further explored the potential variables that could improve the explanation of the variability of the reference scan, when using the alternative procedure. For each model (whole-body and subtotal bone mineral content, fat mass, and lean soft tissue), potential covariates such as age, sex, athletic status, and interactions were not associated with results from the reference scan.

### 4.5. Discussion

Although DXA provides an accurate measurement of body composition, it presents limitations when evaluating taller participants, as whole-body scans cannot be obtained since individuals will be outside the scan area. In order to solve this methodological limitation, the main purpose of this study was to analyse the accuracy of using the sum of two separated scans: head scan and trunk plus limbs scan, to assess bone mineral content, fat mass, and lean soft tissue, using a whole-body scan as the reference criteria. Our results demonstrated that the sum of two partial scans provides an accurate assessment of whole-body values.

In the current investigation, using a diverse sample of male and female athletes and non-athletes, we observed that, a) the proposed alternative procedure for fat mass and lean soft tissue did not differ from the reference scan ( $\mathrm{p}>0.05$ ); b) the proposed models explained more than $99 \%$ of the variation in body composition assessed by the reference scan with low standard errors of estimation; c) high concordance correlation coefficients existed (> 0.99) which indicates an almost perfect strength of agreement [16]; and d) agreement analysis demonstrated low limits of agreement. These results indicate a good accuracy of the alternative method to assess both whole-body and subtotal bone mineral content, fat mass, and lean soft tissue. Moreover, the proposed alternative provided precise measures as observed by the low pure error and percent coefficient of variation within the expected values for DXA measures [7]. It is known that DXA measurements vary slightly by type of soft tissue with lean soft tissue demonstrating a better precision [7]. In accordance, we observed that fat mass presented the higher coefficient of variation while a higher precision was found for lean soft tissue.

To our knowledge, only two previous studies [10, 11] have proposed and validated procedures to assess whole-body bone mineral content, fat mass, and lean soft tissue to evaluate individuals taller than the DXA scan area. Silva et al. [11] compared whole-body composition measurements using the knees bent at a $90^{\circ}$ angle, and predictive calibration equations were developed for bone mineral content, fat mass, and lean soft tissue measurements using Hologic DXA equipment, (QDR-1500, pencil-beam mode Waltham, USA). In the aforementioned study, there were differences between the
two positions, for bone mineral content, fat mass, and lean soft tissue by sex, and the whole sample (P50.001). Therefore, three models were developed to calibrate these body components using the knees-bent position. These models explained $99 \%$ of the variation in whole-body composition with standard errors of estimation of 0.05 kg for bone mineral content, 0.69 kg for fat mass, and 0.72 kg for lean soft tissue. Our values for the standard errors of estimation were lower for these three components.

Despite the fact that our study, using fan-beam equipment, requires two scans, it presents a faster scan and a less demanding protocol, as a goniometer was required by Silva et al. (2004) [11] to establish the correct knees reference position $\left(90^{\circ}\right)$. Moreover we also verified that our procedure is accurate to estimate subtotal body composition (without the head), which would only require one body scan. The use of subtotal results also allows for the same extra height advantage ( $\sim 20 \mathrm{~cm}$ ), while still evaluating limbs and trunk. It is important to highlight that DXA excludes pixels that contain bone in addition to soft tissue for calculating fat mass and lean soft tissue and therefore these values are estimated based on the composition of the adjacent soft tissue pixels [17]. In the head, due to the skull bone, DXA measures are conducted based on this assumption and therefore, in addition to the fact that only one scan would be necessary, subtotal results may present less sources of systematic error in fat mass and lean soft tissue estimations. However, using subtotal values, whole-body bone mineral content is not fully estimated which compromises body composition assessment when using multicomponent models. Molecular multi-component models are widely used in the research setting as they account for more biological variability by partitioning fat-free mass into two or more components (e.g. water, mineral, and protein) [18]. Whole-body body composition assessment may also be useful when considering population reference values [19] and for comparison purposes within a specific sport [6].

Considering that in our investigation the bias of the methods $(0.01 \mathrm{~kg}, 0.06 \mathrm{~kg}$, and 70.07 kg , respectively, for bone mineral content, fat mass, and fat-free mass) were within our technical errors of measurement $(0.03 \mathrm{~kg}$ for bone mineral content, 0.21 kg for fat mass, and 0.34 kg for lean soft tissue), we decided not to develop calibration models for bone mineral content, fat mass, and lean soft tissue when using the sum of two scans. Nevertheless, we analysed the potential variables that could improve the explanation of the variability of the reference scan, when using the alternative
procedure. Since none of these variables (age, sex, athletic status, and interactions) were significant predictors, bone mineral content, fat mass, and lean soft tissue estimated by the reference procedure were only explained by the single sum of two scans. However, it is important to highlight that there is an individual error reflected when using the sum of two scans as the alternative procedure. For instance, the proposed alternative procedure can overestimate fat mass by 0.94 kg or underestimate it by 1.07 kg , given the $95 \%$ limits of agreement.

The other previous study conducted to solve the methodological limitation of assessing participants taller than the DXA scan area was carried out by Evans et al. [10] and involved summing two scans, using the neck and hip as body sites to delimit the scan area. The authors used pencil-beam equipment, Hologic QDR/W 1.000 (Waltham, MA; Enhanced Whole-Body Analysis software version 5.71), and standard errors of estimation values for bone mineral content, fat mass, and lean soft tissue were 0.026 kg , 0.44 kg , and 0.62 kg , respectively, for the hip method, and $0.03 \mathrm{~kg}, 0.28 \mathrm{~kg}$ and 0.33 kg , respectively, for the neck method. Similarly to our results all $r^{2}$ values were higher than 0.99 . Both of these studies [10,11] found a better accuracy for bone mineral content and lean soft tissue, than fat mass measurements. However it is important to underscore that the two previous studies used pencil-beam mode equipment, while our equipment used a fan-beam mode. The fan-beam array distributes the overlapped X-ray across a wider area, shaped like an open fan [20]. The narrower angle fan-beam eliminates beam distortion at the end of a beam path [21], thus, differences in body composition can be observed when DXA pencil- and fan-beam equipments from the same and different manufacturer are compared [21, 22].

Despite the encouraging results obtained in the current study, some limitations should be addressed. Our results are of practical interest to a laboratory with the same model densitometer (Hologic Explorer-W), software, and fan-beam mode. Therefore, our method may not be appropriate for equipment developed by other manufacturers, or using a different software and scan mode. In addition, our validation study was performed in a cross-sectional cohort. It would be useful to establish the validity of the suggested method in longitudinally-monitored populations. Furthermore, our sample comprised young healthy adults that were normal or overweight, consequently our procedure may not be generalized to older and obese populations. Finally, despite the
fact that subtotal body composition results may be used, the whole-body scan procedure requires two scans to assess body components, which would require a longer time. However, in fan-beam densitometers, the time spent for a whole-body scan is considerably shorter, compared to equipment using pencil-beam mode. Both scans can be performed in less than 10 minutes as the length for the head scan can be set to a smaller area considerably reducing the scan time. An extra height of 15 to 20 cm can be gained with this procedure; however the scan area is still limited by the head length that can be dropped off the examination table.

### 4.6. Conclusions

In conclusion, the method proposed is an alternative solution to be used in individuals taller than the DXA scan area, specifically elite athletes engaged in sports recognised for including very tall individuals, such as basketball and volleyball players. Considering the need for obtaining accurate individual body composition measurements throughout the season in elite athletes that are taller than the DXA scan area, the sum of two scans (head and trunk plus limbs) procedure provides a valid and non-invasive approach, allowing the evaluation of participants whose height exceeds the height of the available standard scan by up to 15 to 20 cm .

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# Validity of a combined heart rate and motion sensor for the measurement of free-living energy expenditure in <br> <br> very active individuals ${ }^{2}$ 

 <br> <br> very active individuals ${ }^{2}$}

[^1]
# Validity of a combined heart rate and motion sensor for the measurement of free-living energy expenditure very active individuals 

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### 5.1. Abstract


#### Abstract

Aim: The correct assessment of energy expenditure in very active individuals is important to ensure that dietary energy intake is sufficient. We aimed to validate a combined heart rate (HR) and motion sensor in estimating total (TEE) and activity energy expenditure (AEE) in male and female females with high physical activity levels.


Method: Doubly-labelled water (DLW) was used to assess 7-day TEE in 12 male and female elite junior basketball players, aged 16-17 years. Resting energy expenditure (REE) was assessed with indirect calorimetry and AEE was calculated (AEE $=$ TEE REE - $0.1 \times$ TEE). Simultaneously, TEE and AEE were measured by combined HR and motion sensing. Individual HR calibration was performed with step-test. TEE and AEE were estimated from accelerometry and HR with individual $\left(\mathrm{ACC}+\mathrm{HR}_{\text {step }}\right)$ and group calibration ( $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ ).

Results: Mean differences from AEE from DLW were found when using the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}(-17.7 \mathrm{~kJ} / \mathrm{kg} / \mathrm{day})$. The combined sensor results were correlated with TEE (kJ/day) $\quad\left[r^{2}=0.53 \quad\left(\mathrm{ACC}+\mathrm{HR}_{\text {step }}\right) ; \quad \mathrm{r}^{2}=0.57 \quad\left(\mathrm{ACC}+\mathrm{HR}_{\text {group }}\right)\right]$ and $\mathrm{AEE} \quad\left[\mathrm{r}^{2}=0.21\right.$ $\left.\left(\mathrm{ACC}+\mathrm{HR}_{\text {step }}\right) ; \mathrm{r}^{2}=0.21\left(\mathrm{ACC}+\mathrm{HR}_{\text {group }}\right)\right]$ from DLW though no association was found for relative energy expenditure ( $\mathrm{EE} / \mathrm{kg}$ ). Higher coefficients of determinant were observed when considering the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ instead of the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$. Higher CCC values were observed for the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ ( $\sim 0.5$ for relative TEE and AEE). Large limits of agreement were found $\left[\mathrm{ACC}+\mathrm{HR}_{\text {step }}:-70,48\right.$ (TEE) and $-77,42$ (AEE) $\mathrm{kJ} / \mathrm{kg} / \mathrm{day}$; $\mathrm{ACC}+\mathrm{HR}_{\text {group: }}$ : $-71,64$ (TEE) and -67 , 45 (AEE) $\left.\mathrm{kJ} / \mathrm{kg} / \mathrm{day}\right]$.

Conclusions: ACC+HR models are a valid alternative to estimate TEE but not AEE for a group of very active individuals. However, the combined monitor is not accurate to assess individual energy requirements.

Key Words: athletes; energy requirements; doubly labeled water; indirect calorimetry; heart rate monitoring; accelerometry.

### 5.2. Introduction

Physical activity energy expenditure (AEE) is the most variable component of total energy expenditure (TEE). In very active individuals daily TEE can be twice as much as resting energy expenditure [1], and during heavy sustained exercise in the Tour de France a fivefold increase has been described [2]. With limited energy intake, lean tissue will be used as fuel resulting in loss of strength and endurance that may compromise immune, endocrine, and musculoskeletal function.[3] Energy deficient females can develop a cluster of conditions named "female athlete triad", leading to amenorrhea, osteopenia, and premature osteoporosis, among others [4]. Very active individuals are more likely to be chronically energy deficient, thus it is important to precisely measure energy expenditure (EE) to identify individual energy requirements [5].

Doubly labelled water (DLW) is the gold standard to assess TEE in free-living individuals and it has frequently been used in highly trained athletes [6, 7], However the analytical procedures involved in dilution techniques are time-consuming, expensive, and involve complex methods and specialized technicians, excluding its routine use for EE assessment [8]. Other alternative, objective, and valid methods to assess EE need to be validated in a population with high levels of EE.

Motion sensors and heart rate (HR) monitors provide objective measures of EE, however both present limitations. Motion sensors, worn on the hip are not capable of detecting upper body movements, changes in grade during walking and running, and free weight exercises [9], and evidence exists that the relation between accelerometry and physical activity intensity (PAI) is affected at higher intensities [10, 11]. For partially solving this problem, other wearing locations have been proposed, especially at the ankle and wrist placements. However, limb-worn motion sensors provide similar EE
outcome values as the hip-worn motion sensors, during free-living conditions. [12] In active individuals HR is often used as a physiological objective variable, directly associated with oxygen consumption [13, 14], though the association between EE and HR can be influenced by other factors [14, 15]. Moreover HR does not present a good accuracy in estimating EE of individuals with high physical activity levels [14, 16]. The use of both methods combined may provide more accurate measures of EE [17, 18]. A monitor combining HR and accelerometry into a single device has been developed [18] and validated [19-21], though its validity in very active individuals has not been examined.

The aim of our investigation was to assess the validity of a combined HR and motion sensor in estimating free-living TEE and AEE in very active males and females using DLW as the reference method.

### 5.3. Methods

### 5.3.1. PARTICIPANTS

Twelve male and 12 female basketball players from the Portuguese Junior National Team volunteered to participate, however only 12 participants had valid records of the combined HR and motion sensor, therefore 4 males ( 2 guards and 2 forwards) and 8 females ( 2 guards, 4 forwards and 2 centers) were used in this study. Therefore, 8 males and 4 females were excluded for not having valid records of the combined monitor.

### 5.3.2. EXPERIMENTAL DESIGN

Energy expenditure of the participants was evaluated at the first or second week of the pre-season training period (September) during a 7-day period. The players lived and trained at the National High-Performance Center during the week days. In the end of the week athletes went to their homes and trained with their respective teams on Friday and Saturday. On Sunday afternoon the athletes came back to the training center. The male and female training regimens while in the training center consisted of 4
technical-tactical 120 minutes sessions (1/day), and resistance training for 60 minutes two times during the week. In addition players participated in one training game in the middle of the week. Apart from the training regimens, athletes went to school every week day and had two 90 minute physical education classes during the period of assessment.

Inclusion criteria were: 1) Tanner stage V [22]; 2) $\geq 10$ hours training/week; 3) negative test outcomes for performance-enhancing drugs; and 4) not taking any medications or dietary supplements. No females were taking oral contraceptives. Medical screening indicated that all subjects were in good health, without endocrine abnormalities that would limit their participation in the study. All subjects and guardians were informed about the possible risks of the investigation before giving written informed consent to participate. All procedures were approved by the Ethics Committee of the Faculty of Human Kinetics, Technical University of Lisbon, and conducted according to the declaration of Helsinki for human studies of the World Medical Association.

Subjects came to the laboratory in a 12 h fasted state and consumed a normal evening meal the night before testing. Vigorous exercise was not allowed for at least 14 h and caffeine, alcohol or stimulant beverages for at least 24 -hours before testing begin at 08:00 a.m.

### 5.3.3. BODY COMPOSITION MEASUREMENTS

Body weight and height were measured to the nearest 0.01 kg and 0.1 cm , respectively, according to standardized procedures [23].

Dual energy X-ray absorptiometry (Hologic Explorer-W, software QDR for Windows v.12.4, Waltham, Massachusetts, U.S.A.) was used to estimate fat mass (FM) and fat-free mass (FFM) [24]. Hologic fan-beam densitometers provide valid body composition estimates in athletes [25] The coefficient of variation (CV) in our laboratory, based on 10 young active adults (five males and five females) for FM and FFM are $1.7 \%$ and $0.8 \%$, respectively [26].

### 5.3.4. Energy Expenditure Measurements

## Resting energy expenditure

Resting energy expenditure (REE) measurements were performed by an opencircuit indirect calorimetry through a portable gas analyser ( $\mathrm{K} 4 \mathrm{~b}_{2}$, Cosmed, Rome, Italy) while participants were lied supine wearing a with a face mask for data collection, as described elsewhere [7]. For data analysis a steady state was defined as a 5 -min period with $\leq 10 \% \mathrm{CV}$ for $\dot{\mathrm{V}} \mathrm{O}_{2}$ and $\dot{\mathrm{V}} \mathrm{CO}_{2}$ [27]. The mean $\dot{\mathrm{V}} \mathrm{O}_{2}$ and $\dot{\mathrm{V}} \mathrm{CO}_{2}$ of a 5 -min steady states were used in Weir equation [28] and the period with the lowest REE was considered. The CV for REE in our laboratory is $10 \%$.

## Total energy expenditure from doubly labelled water

Total energy expenditure was measured during a 7-day period by an established procedure using deuterium oxide and 18-Oxygen. An oral dose of $0.8 \mathrm{~g} / \mathrm{kg}$ of total-body water (TBW) of $\approx 10$ atom $\%(A P) \mathrm{H}_{2}{ }^{18} \mathrm{O}$ (Taiyo Nippon Sanso Corporation, Tokyo, Japan), assuming TBW is $61 \%$ of body mass, and $0.16 \mathrm{~g} / \mathrm{kg}$ of TBW of $99.9 \mathrm{AP}^{2} \mathrm{H}_{2} \mathrm{O}$ (Sigma-Aldrich, Co, St Louis, Mo, USA), diluted in 50 ml of water was administered to the subjects. The analytical procedures used to estimate TEE are described elsewhere [7]. The CV for TEE is $4.3 \%$. AEE was calculated as TEE - REE $-0.1 \times$ TEE, assuming the thermic effect of food is $\sim 10 \%$ of TEE) and physical activity level (PAL) was determined as TEE/REE.

## Total energy expenditure from combined heart rate and motion sensor

Energy expenditure simultaneously also evaluated with combined HR and motion sensor (Actiheart, Cambridge Neurotechnology Ltd, Cambridge, UK). The monitor was worn using ECG pads in the chest during the same 7-day period that the DLW assessment took place. Participants performed an 8-min step-test (height: 215 mm ), the stepping speed ramps linearly increased from 15-33 step cycles/min, providing individual HR-EE relationship calibration. From the individual step-test calibration estimated $\mathrm{VO}_{2}$ max was derived by the software. The device was started with 60-s epochs and participants were asked to wear the monitor at all times (even during sleep hours) for the 7 -consecutive days the DLW assessment were taking place. Data
from the monitors were downloaded into to the commercial software (v.4.0.46). The software algorithm allowed data cleaning, recovering, and interpolation of missing and noisy HR. Only participants with 3 -valid days were considered for data analysis. A valid day was considered when we had at least $70 \%$ of the day ( 1008 min ) with records and not more than $10 \%$ of the registered timed with HR recovered by the software. Moreover, if the participants had invalid data during the training hours (registered in a diary) the day was not considered valid.AEE was estimated using energy models, available in the commercial software:
$\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ : individual HR calibration model (Group CalJAP2007/Step HR [29]), with HR and accelerometry data;

ACC $+\mathrm{HR}_{\text {group }}$ : group HR calibration model(Group CalJAP2007 [29]) with HR and accelerometry data;
$\mathrm{HR}_{\text {flex }}$ : individual HR calibration model (Group CalJAP2007/Step HR [29]), with HR data;

ACC: accelerometry data [29].
TEE was estimated adding to the estimated AEE, the thermic effect of food ( $\sim 10 \%$ of TEE) and REE using the Schofield equation,[30] as suggested by the manufacturer.

### 5.3.5. MAXIMAL OXYGEN CONSUMPTION

Maximal oxygen consumption $\left(\mathrm{VO}_{2 \max }\right)$ measurement was performed with a continuous, progressive treadmill running protocol in a laboratory $\left(21-22^{\circ} \mathrm{C}\right.$, relative humidity of $50 \%$ ). Following a $2-\mathrm{min}$ warm-up (males: $0 \%$ grade; $7 \mathrm{~km} / \mathrm{h}$ speed; females: $0 \%$ grade; $6 \mathrm{~km} / \mathrm{h}$ speed), subjects ran for $2-\mathrm{min}$ (males: $0 \%$ grade; $9 \mathrm{~km} / \mathrm{h}$ speed; males: $0 \%$ grade; $8 \mathrm{~km} / \mathrm{h}$ speed). Speed and grade were incremented $1 \mathrm{~km} / \mathrm{h}$ and $1 \%$, respectively, every $2-\mathrm{min}$, until exhaustion. Subjects received verbal encouragement and where instructed to exercise to volitional fatigue. Breath-by-breath gases were continuously analyzed with an open-circuit spirometry system (Quark $\mathrm{b}^{2}$, Cosmed, Rome, Italy). Heart rate was continuously measured during the test (Polar Electro Oy, Kempele, Finland). $\mathrm{VO}_{2 \max }$ was attained when at least two of the following
three criteria were achieved: no increase in $\mathrm{VO}_{2 \max }$ despite further increases in work rate, a heart rate at or above age predicted maximum, and/or a RER $\geq 1.0$.

### 5.3.6. STATISTICAL ANALYSIS

Statistical analysis was performed using IBM-SPSS Statistics v.19.0 (SPSSIBM, Chicago, Illinois, U.S.A.) and the MedCalc Statistical Software v.11.1.1.0 (Mariakerke, Belgium). Descriptive statistics were calculated for all outcome measurements. Normality was tested using Shapiro-Wilk test. Comparisons between sexes were performed using independent sample t-test or the Mann-Whitney U test. Because no sex by EE (combined HR and motion sensor) was observed all analysis were performed using the whole sample. Methods comparisons were performed using paired sample T-test or Wilcoxon-test. Simple linear regressions were performed to calculate the relationship between EE from DLW and the combined sensing. The concordance coefficient correlation (CCC) was analyzed to evaluate the degree to which pairs of observations fall on the $45^{\circ}$ line through the origin. The CCC $\left(\rho_{c}\right)$ contains a measurement of precision $\rho$ and accuracy ( $\rho_{\mathrm{c}}=\rho \mathrm{C}_{\mathrm{b}}$ ) where $\rho$ is the Pearson correlation coefficient, which measures how far each observation deviates from the best-fit line, and is a measure of precision, and $\mathrm{C}_{\mathrm{b}}$ is a bias correction factor that measures how far the best-fit line deviates from the $45^{\circ}$ line through the origin, and is a measure of accuracy. Agreement between methods was assessed [31], including the $95 \%$ limits of agreement (LoA). The correlation between the mean and the difference of both methods was used as an indication of proportional bias. For all tests significance was set at $\mathrm{p}<0.05$.

### 5.4. Results

Participant's characteristics and descriptive statistics for $\mathrm{VO}_{2 \text { max }}$, REE, TEE, and AEE are summarized in table 5.1.

Table 5.1. Descriptive statistics and data from the combined HR and motion sensor monitor and from doubly-labelled water (results are expressed as mean $\pm$ SD).

|  | All sample $\mathrm{n}=12$ | Males $n=4$ | Females $n=8$ |
| :---: | :---: | :---: | :---: |
| Age (years) | $16.4 \pm 0.5$ | $16.5 \pm 0.6$ | $16.4 \pm 0.5$ |
| Weight (kg)* | $67.7 \pm 8.6$ | $74.5 \pm 6.5$ | $64.3 \pm 7.6$ |
| Height (cm)* | $180.6 \pm 7.8$ | $189.9 \pm 2.8$ | $175.9 \pm 4.3$ |
| BMI (kg/m ${ }^{2}$ ) | $20.8 \pm 2.4$ | $20.7 \pm 1.7$ | $20.8 \pm 2.7$ |
| \%FM ${ }^{*}$ | $20.0 \pm 5.6$ | $13.8 \pm 2.6$ | $23.0 \pm 3.6$ |
| FM (Kg) ${ }^{\text { }}$ | $13.5 \pm 3.6$ | $9.5 \pm 1.1$ | $15.5 \pm 2.6$ |
| FFM (Kg) ${ }^{*}$ | $53.9 \pm 8.2$ | $63.3 \pm 4.9$ | $49.2 \pm 4.7$ |
| Sleep HR (beats/min) ${ }^{*}$ | $50.4 \pm 6.0$ | $45.5 \pm 4.7$ | $52.9 \pm 5.2$ |
| PAL from DLW | $2.35 \pm 0.52$ | $2.37 \pm 0.69$ | $2.34 \pm 0.46$ |
| $\mathrm{VO}_{2 \text { max Treadmill }}\left(\mathrm{ml} / \mathrm{kg} / \mathrm{min}\right.$ ) ${ }^{*}$ | $56.9 \pm 6.7$ | $63.3 \pm 1.6$ | $53.7 \pm 5.7$ |
| $\mathrm{VO}_{2 \text { max Step test }}(\mathrm{ml} / \mathrm{kg} / \mathrm{min}){ }^{* \#}$ | $45.0 \pm 5.3$ | $51.5 \pm 2.3$ | $41.7 \pm 2.2$ |
| $\mathrm{REE}_{\text {indirect calorimetry }}(\mathrm{kJ} /$ day $)$ | $6510 \pm 991$ | $7277 \pm 1023$ | $6127 \pm 769$ |
| REE Schofield $^{(k J / d a y)}{ }^{\text {* }}$ * | $7115 \pm 946$ | $8320 \pm 446$ | $6512 \pm 278$ |
| TEE from DLW (kJ/day) | $15059 \pm 2864$ | $16762 \pm 3070$ | $14208 \pm 2523$ |
| TEE from Actiheart ${ }^{\text {a }}$ ( $\mathrm{kJ} /$ day) |  |  |  |
| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}{ }^{*}$ | $14349 \pm 2402$ | $16696 \pm 2845$ | $13175 \pm 935$ |
| $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ | $14914 \pm 3534$ | $18285 \pm 4290$ | $13229 \pm 1413$ |
| $\mathrm{HR}_{\text {flex }}{ }^{\text {*\# }}$ | $17866 \pm 3147$ | $20729 \pm 3955$ | $16434 \pm 1352$ |
| ACC ${ }^{* \#}$ | $11406 \pm 1730$ | $13201 \pm 1732$ | $10508 \pm 935$ |
| TEE from DLW (kJ/kg/day) | $223 \pm 34$ | $224 \pm 34$ | $222 \pm 36$ |
| TEE from Actiheart ${ }^{\text {a) }}$ (kJ/kg/day) |  |  |  |
| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ | $212 \pm 18$ | $223 \pm 23$ | $206 \pm 13$ |
| $\mathrm{ACC}+\mathrm{HR}^{\text {group }}$ | $219 \pm 32$ | $244 \pm 42$ | $207 \pm 18$ |
| $H \mathrm{R}_{\text {flex }}{ }^{*}{ }^{\text {\# }}$ | $264 \pm 26$ | $277 \pm 34$ | $257 \pm 20$ |
| ACC ${ }^{\text {\# }}$ | $168 \pm 12$ | $177 \pm 11$ | $164 \pm 10$ |
| AEE from DLW (kJ/day) | $7043 \pm 2663$ | $7908 \pm 3572$ | $6660 \pm 2275$ |
| AEE from Actiheart ${ }^{\text {a) }}$ (kJ/day) |  |  |  |
| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ | $5799 \pm 1409$ | $6706 \pm 2198$ | $5346 \pm 586$ |
| $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ | $6308 \pm 2432$ | $8136 \pm 3507$ | $5394 \pm 1075$ |
| $\mathrm{HR}_{\text {flex }}{ }^{\text {\# }}$ | $8964 \pm 2113$ | $10335 \pm 3209$ | $8278 \pm 994$ |
| ACC ${ }^{\text {\# }}$ | $3150 \pm 794$ | $3560 \pm 1166$ | $2945 \pm 513$ |
| AEE from DLW (kJ/kg/day) | $103 \pm 34$ | $103 \pm 43$ | $103 \pm 32$ |
| AEE from Actiheart ${ }^{\text {a) }}$ (kJ/kg/day) |  |  |  |
| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ | $85 \pm 13$ | $89 \pm 23$ | $83 \pm 6$ |
| $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ | $92 \pm 27$ | $107 \pm 40$ | $84 \pm 14$ |
| $\mathrm{HR}_{\text {flex }}{ }^{\text {\# }}$ | $132 \pm 21$ | $137 \pm 33$ | $129 \pm 13$ |
| ACC ${ }^{\text {\# }}$ | $46 \pm 7$ | $47 \pm 12$ | $46 \pm 5$ |

Significant differences between sexes
\#Significantly different from the reference method (all sample)
*Models of energy expenditure prediction from Actiheart: $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ - individual HR calibration model (Group Cal JAP2007/Step HR, with HR and accelerometry data; ACC $+\mathrm{HR}_{\text {group }}$ - group HR calibration model (Group Cal JAP2007) with HR and accelerometry data; $\mathrm{HR}_{\text {flex }}$ - using the individual HR calibration model (Group Cal JAP2007/Step HR) with HR data; ACC: using accelerometry data.
Abbreviations: BMI, body mass index; FM, fat mass; FFM, fat-free mass; HR, heart rate; PAL, physical activity level; $\mathrm{VO}_{2}$ max, maximal oxygen consumption; REE, resting energy expenditure; TEE, total energy expenditure; DLW, doubly labelled water; AEE, activity energy expenditure.

Table 5.2 presents the validity of the combined HR and motion sensor in estimating EE from the reference method.

Table 5.2. Validity of the energy expenditure models from the combined heart rate and motion sensor monitor.

| Regression <br> analysis | CCC analysis | Agreement analysis |  |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| $\mathrm{r}^{2} \quad$ see | CCC | $\rho$ | $\mathrm{C}_{\mathrm{b}}$ | Bias $95 \%$ LoA | Trend |

Total Energy Expenditure (kJ/day) ${ }^{\text {a) }}$

| ACC $+\mathrm{HR}_{\text {step }}$ | $\mathbf{0 . 5 3}^{*}$ | 2070 | 0.69 | 0.7246 | 0.9474 | -710 | $-4632 ; 3211$ | $-0.25(\mathrm{p}=0.436)$ |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ | $\mathbf{0 . 5 7}^{*}$ | 1964 | 0.74 | 0.7567 | 0.9772 | -145 | $-4689 ; 4398$ | $0.31(\mathrm{p}=0.330)$ |
| $\mathrm{HR}_{\text {flex }}$ | $\mathbf{0 . 4 9}^{*}$ | 2147 | 0.47 | 0.6995 | 0.6752 | 2806 | $-1788 ; 7401$ | $0.13(\mathrm{p}=0.686)$ |
| ACC | $\mathbf{0 . 4 4}^{*}$ | 2252 | 0.25 | 0.6617 | 0.3847 | -3654 | $-7875 ; 568$ | $-0.57(\mathrm{p}=0.051)$ |

Total Energy Expenditure ( $\mathrm{kJ} / \mathrm{kg} / \mathrm{day})^{\text {a) }}$

| ACC $+H R_{\text {step }}$ | 0.21 | 31.5 | 0.35 | 0.4552 | 0.7708 | -10.8 | $-70.0 ; 48.4$ | $-0.59(p=0.043)$ |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| ACC $+H R_{\text {group }}$ | 0.21 | 31.5 | 0.45 | 0.4556 | 0.9918 | -3.7 | $-70.9 ; 63.6$ | $-0.06(p=0.848)$ |
| HR $_{\text {flex }}$ | 0.20 | 31.7 | 0.21 | 0.4446 | 0.4809 | 41.1 | $-22.0 ; 104.2$ | $-0.28(p=0.379)$ |
| ACC | 0.06 | 34.4 | 0.04 | 0.2415 | 0.1744 | -54.2 | $-118.8 ; 10.4$ | $-0.80(p=0.002)$ |

Activity Energy Expenditure(kJ/day) ${ }^{\text {a) }}$

| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ | $0.41{ }^{*}$ | 2141 | 0.45 | 0.6424 | 0.6972 | -124 | 5287; 2800 | ) |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| ACC + HR group | 0.53* | 1922 | 0.69 | 0.7256 | 0.9527 | -735 | -4458; 2987 | 4) |
| $\mathrm{HR}_{\text {flex }}$ | 0.33 | 2401 | 0.41 | 0.5720 | 0.7221 | 1921 | -2514; 6356 | -0.27 ( $p=0.389$ ) |
| ACC | 0.31 | 2324 | 0.10 | 0.5547 | 0.1743 | -3893 | -8438; 653 | 0.88 ( $\mathrm{p}<0.001$ ) |

Activity Energy Expenditure(kJ $/ \mathrm{kg} /$ day $)^{\text {a) }}$

| $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ | 0.19 | 31.8 | 0.24 | 0.4366 | 0.5447 | -17.7 | $-77.2 ; 41.9$ | $-0.76(p=0.004)$ |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| $A C C+H R_{\text {group }}$ | 0.33 | 29.0 | 0.52 | 0.5714 | 0.9048 | -11.2 | $-67.4 ; 44.9$ | $-0.28(p=0.375)$ |
| $\mathrm{HR}_{\text {flex }}$ | 0.11 | 33.4 | 0.19 | 0.3309 | 0.5591 | 29.1 | $-36.0 ; 94.1$ | $-0.48(p=0.115)$ |
| $A C C$ | 0.06 | 34.3 | 0.03 | 0.2464 | 0.1073 | -56.7 | $-120.8 ; 7.4$ | $-0.91(p<0.001)$ |

a)Models of energy expenditure prediction from Actiheart: $A C C+H R_{\text {step }}$ - individual HR calibration model (Group Cal JAP2007/Step HR, with HR and accelerometry data; ACC+HR group - group HR calibration model (Group Cal JAP2007) with $H R$ and accelerometry data; $\mathrm{HR}_{\text {flex }}$ - using the individual HR calibration model (Group Cal JAP2007/Step HR) with HR data; ACC: using accelerometry data.
*Significant associations ( $p<0.05$ )

Absolute EE from the combined sensing were related to the results from DLW using combined HR and motion sensor, however no significant associations were verified when estimating AEE using the $H R_{\text {flex }}$ and the ACC models. The combined HR and motion sensor models explained between $44 \%$ (ACC) and $57 \%$ (ACC+HR group) of absolute TEE by DLW. For absolute AEE, the explained variance was lower
corresponding to $41 \%$ for the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ and $53 \%$ for the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ model. When analysing the accuracy of relative values (EE/kg) lower coefficients were found with no significant associations between the alternative and the reference methods. The higher CCC was obtained for the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$, both for absolute TEE and AEE. Lower CCC were observed when analysing the overall accuracy and precision of the combined monitor in assessing EE/kg.


Abbreviations: TEE, total energy expenditure; AEE, activity energy expenditure; ACC $+\mathrm{HR}_{\text {step }}$, model with accelerometry and individual step test calibration of heart rate; $A C C+H R_{\text {group }}$, model with accelerometry and group calibration of heart rate.
Figure 5.1. Bland-Altman analysis of the agreement between methods in assessing total and activity energy expenditure using branched equation models, using the individual HR calibration (ACC+HRstep) or using the group HR calibration (ACC+HRgroup).

On an individual level, the best LoA were established for the relative TEE obtained from $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ (Table 5.2), with an individual error between an underestimation of $70 \mathrm{~kJ} / \mathrm{kg} /$ day to an overestimation of $48 \mathrm{~kJ} / \mathrm{kg} / \mathrm{day}$, though a significant trend between the mean and the difference of the methods was found. For relative AEE the best LoA were observed for the ACC+HR group model (table 5.2), where
the individual error can be underestimated by $67 \mathrm{~kJ} / \mathrm{kg} /$ day or overestimated by 45 $\mathrm{kJ} / \mathrm{kg} / \mathrm{day}$. In Figure 5.1 are illustrated the Bland-Altman plots for the combined HR and motion sensor models that consider the combined HR and ACC models $\left(\mathrm{ACC}+\mathrm{HR}_{\text {step }}\right.$ and the $\left.\mathrm{ACC}+\mathrm{HR}_{\text {group }}\right)$.

### 5.5. Discussion

To our knowledge this is the first study to examine the validity of a combined HR and motion sensor in assessing EE in young elite athletes using DLW as the reference criteria.

In this investigation we verified that the results from the combined measures of both HR and accelerometry did not differ from the DLW method. However estimation of both AEE and TEE were underestimated by the ACC model and overestimated by the $\mathrm{HR}_{\text {flex }}$ model, respectively. Moreover, the results from all models were associated with absolute TEE from the DLW, despite the models that only used one measure (accelerometry or HR) did not explain the DLW absolute AEE results. On the other hand, relative EE results (EE/kg body weight) were not significantly associated with DLW values.

Several investigations have validated combined HR and motion sensors but in different populations [32, 33]. However, to our knowledge, so far only two studies assessed the validity of the monitor with the Cambridge Neurotechnology algorithms (Actiheart, CamNtech Limited, UK) in free-living conditions using DLW as the reference criterion [20, 21]. Assah et al. [20] observed a significantly higher relative AEE in rural compared to urban participants, reporting higher associations between methods for urban individuals, ranging from 0.40 ( $\mathrm{HR}_{\text {flex }}$ ) to 0.70 (ACC) in urban and from $0.25\left(\mathrm{HR}_{\text {flex }}\right)$ to 0.45 in rural (ACC). In our investigation, when using relative EE (EE/kg), non-significant associations were observed between TEE and AEE from the combined sensor with the reference results from DLW [r: 0.2-0.5 (TEE) and 0.2-0.6 (AEE)].

Total and activity energy expenditure from the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ were overall more accurate and precise than TEE and AEE from the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ in estimating results from DLW. These findings do not fully extend those reported by Villars et al. [21], as
the individual calibration was more accurate than the group calibration. It is important to note that Villars et al. [21], used a specific graded maximal test to perform the individual calibration and did not rely on the available calibration test provided by the equipment which may have lead to lower individual errors given by the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ model. The combined HR and motion sensor commercial software includes a built-in step-test protocol that is used for deriving the individual HR-EE relationship in the field, and EE is then calculated $\left(\mathrm{ACC}+\mathrm{HR}_{\text {step }}\right)$. When the step-test is not performed it is possible to select a model that uses a group calibration, which is an approximation for a range of individual fitness levels [29], Assah et al. [20] observed that this individual step-test calibration did not bring an improved validity to the group calibration model. It is important to highlight that even when using the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$, although individual PAI-HR curves are not considered for EE calculations, the model considers HR above sleep (HRas) and sex for AEE calculations. Brage et al. [29] reported that some of the HR variance can be accounted for simply using HRas instead of HR and adjusting for sex.

In our study we found that the mean $\mathrm{VO}_{2 \text { max }}$ was underestimated by the step-test (Table 5.1), comparing to the treadmill test with gas analysers. In fact, a potential explanation for the unexpected less valid AEE results by using the individual compared to the group calibration model is the lack of accuracy of the step-test in deriving the individual HR-PAI relationship in participants that perform daily exercise at higher levels of intensity. Indeed higher standard errors of estimation (SEE) were observed, specifically for the relative values of AEE ranging from $29.0 \mathrm{~kJ} / \mathrm{kg} / \mathrm{day}$ ( $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ ) to $34.3 \mathrm{~kJ} / \mathrm{kg} / \mathrm{day}\left(\mathrm{HR}_{\mathrm{flex}}\right)$. Further research is required to provide an alternative field protocol for the HR-EE individual calibration in elite athletes.

Agreement analysis demonstrated a significant trend between the mean and the difference of the methods in estimating relative EE for the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$, and the ACC model, meaning that the individual errors from combined HR and motion sensor is dependent on the individual EE, when using these models. These results agree with previous observations [20] that TEE and AEE from the combined sensor appear less accurate with increasing AEE. Considering the large LoA observed for all models our results revealed large individual estimation errors. The lowest LoA for TEE was observed for the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ model, whereas for AEE the lowest individual error was
found for the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ model. Contrarily, Villars et al. [21] observed lower LoA in estimating AEE when using an individual calibration; however the authors used an exercise tolerance test, and not the step-test to improve AEE estimations.

Accelerometry models provided the highest individual errors in the present investigation. Accelerometry, being a biomechanical measure of movement is expected to be associated with AEE. In our sample the ACC model was the least accurate, underestimating EE which is in accordance with Brage et al. [29]. The authors [29] stated that accelerometry models tend to underestimate PAI, mainly due to the variability of the sources of movement and the assumptions about the efficiency of the work performed. In opposite, Assah et al. [20] verified that the ACC model presented the stronger associations with AEE from DLW compared to other EE models. The mean PAL of our sample was 2.35 , representing a PAL compatible with very active individuals [34]. It is expected that an underestimation of EE may occur when using uni-axial accelerometers as the linearity between counts and aerobic intensity is not always assumed at moderate to high velocities [35]. Nevertheless accelerometers are limited in assessing EE of weight-bearing activities [9], that are part of the training regimens of these basketball players. It has also been demonstrated that the actiheart accelerometer component presents a poor performance compared to other hip equipment, particularly at higher intensities during level walking and level jogging [36]. Therefore the models that only consider accelerometer data may present limited accuracy in specific activities, as the location of the accelerometer component on the sternum may be problematic [37], however these investigations did not focus on the actiheart equipment from the Cambridge Neurotechnology.

The use of HR is not error free as its relation with PAI may be affected by several factors $[14,15]$. It is then expected that EE models that consider both accelerometry and HR data will present better accuracy [10]. For this sample of highly trained athletes, we observed that combining both ACC and HR improved the accuracy of the AEE and TEE estimation by the combined HR and motion sensor, extending the findings of Villars et al. [21]. The authors [21] observed that AEE estimates based on both recordings combined in a weighed branched model, specifically the individual calibration model, correlated better with DLW measures in free-living conditions than estimates from HR or accelerometry alone.

The Actiheart clips on the chest by using standard ECG electrodes that need good skin contact for optimal signal detection. These electrodes may become loose, resulting in noisy or loss of signal detection [20]. In our investigation we observed that, from the 24 participants assessed only 12 ( $50 \%$ ) presented valid records for the combined sensor. This was a problem in our study given that the participants were athletes engaged in high intensity and volume regimen trainings, and as a result the electrodes often lost contact easily due to profuse sweating. This limitation may exclude the combined HR and motion sensor for habitual EE assessment in individuals engaged in high levels of physical activity

## Study limitations.

There are a few limitations in this study. The low number of participants is a potential limitation, as this study is only $80 \%$ powered to detect a correlation coefficient higher than 0.7 in a study with 12 individuals. However, we assessed the entire national junior basketball male and female teams and the results are based on players with valid data. It was not possible to have more valid records since the ECG electrodes needs good skin contact for signal detection and during basketball practices, due to suet, the electrodes often lost contact with the skin loosing HR signal for long periods.

### 5.6. Conclusions

Combine measures of ACC and HR represent a valid alternative to estimate TEE but not and AEE in a very active population, specifically in a group of basketball players at a pre-season that is normally characterized by high-intensity training. Regardless, given the high drop-out rate due to invalid records, concerns may exist when estimating EE from a combined HR and motion sensor. Likewise, considering the wide limits of agreement, the equipment is not accurate to estimate individual energy requirements. We also observed a better accuracy in estimating TEE with the $\mathrm{ACC}+\mathrm{HR}_{\text {group }}$ in opposition to the $\mathrm{ACC}+\mathrm{HR}_{\text {step }}$ model. Further research is needed to test the usefulness of the step-test individual HR-EE calibration incorporated in the combined HR and motion sensor for estimating EE in a population with high levels of physical activity.

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# Association of an entire season with body composition in elite junior basketball players ${ }^{3}$ 

[^2]
# Association of an entire season with body composition in elite junior basketball players 

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### 6.1. Abstract

Aim. Body composition changes among elite athletes may influence competitive performance. This study aimed to characterize the body composition changes at the molecular, cellular, tissue-system, and whole body level of analysis in elite junior basketball players during the course of a season.

Methods. Twelve males and 11 females ( 16 to 17 years) were evaluated. Dualenergy X-ray absorptiometry (DXA) was used to assess bone mineral (Mo) and leansoft tissue (LST). Total-body water (TBW) and extracellular water (ECW) were assessed using isotope dilution techniques, and extracellular (ECF) and intracellular fluids (ICF) were calculated. Fat mass (FM) and fat-free mass (FFM) were assessed with a four-component model. Body cell mass was calculated (LST - (ECF + ECS)). Skeletal muscle (SM) was estimated using appendicular LST (ALST) as: (1.19 $\times$ ALST) - 1.65. At the whole body level, body mass, sum of 7 skinfolds, and muscle circumferences (Mc) were measured. The handgrip and the countermovement jump tests were used for performance assessment.

Results. Males increased FFM ( $4.4 \pm 2.3 \%$ ), TBW ( $3.5 \pm 4.6 \%$ ), SM ( $4.5 \pm$ $2.3 \%)$, and $\operatorname{arm}(3.4 \pm 2.7 \%)$ and thigh $(3.8 \pm 3.0 \%)$ Mc. Females increased SM (5.9 $\pm$ $4.6 \%$ ) and arm ( $3.6 \pm 3.8 \%$ ) and thigh ( $4.0 \pm 5.2 \%$ ) muscle circumferences and decreased ICF ( $-9.7 \pm 13.6 \%$ ). FFM components differed from the established values based on cadaver analysis. Both sexes increased their performance and associations were found between changes in molecular and whole body components with performance.

Conclusion. In conclusion the season was associated with an improved body composition profile in males and few changes in females.

Key words: athletes; body composition; dilution techniques; follow-up study

### 6.2. Introduction

Basketball has achieved an impressive level of popularity in the world today, with 213 National Federations of basketball throughout the world [1]. In the 2009-10 Portuguese League of Basketball regular season, the mean difference among winning and losing a game was only $\sim 11$ points [2]. This small edge underscores the importance of even small increments in performance [3].

Body composition assessment in athletes may help to optimize competitive performance and assess the effects of training and hence is of considerable interest [4, 5]. Several studies developed with elite athletic populations, have reported that an improved body composition may have a positive impact on performance parameters such as maximal oxygen consumption [6], the onset of blood lactate accumulation [6], maximal strength $[7,8]$, and muscle power $[7,8]$.

Changes in body components and physical performance occur from the start to the end of a competitive season in basketball [9-12]. A comprehensive model of human body composition consists of five distinct levels, i.e., atomic, molecular, cellular, tissuesystem, and whole body [13]. Regarding basketball players, the scientific community has been interested mainly in the whole body $[14,15]$ and molecular levels of body composition analysis [10, 12]. Even when assessing molecular components, coaches and investigators tend to pay attention only to fat mass (FM) and fat-free mass (FFM) [ 16,17 ] using two-component (2C) models, which assume that the main components of FFM (mineral, water, and protein) are relatively stable, increasing the chance for errors in body composition assessment [18]. Many coaches and scientists working with elite athletes recognize that, in addition to FM, the knowledge of the amount and distribution of lean tissues can be just as important in determining sports performance [19]. To our knowledge, few studies have characterized body composition in basketball players during the course of a season and all focused on 2 C -based models $[10,12,15]$.

The main goal of a training season is to increase player's performances in the competitions. The greatest fitness improvement occurs in the pre-season, and is normally maintained or may slightly decrease during the in-season period. Body composition assessment is a valuable tool that can help coaches and sports scientists assess and monitor the success of training programs [3, 20]. However, estimates of the
effects of training on body composition are diverse, in part because different assessment techniques of varying accuracy and precision are used to quantify exercise-related changes in body composition [4].

The purpose of the present study was to characterize 4 distinct levels of body composition, molecular, cellular, tissue-system, and whole body, of elite junior male and female basketball players, from the beginning of the pre-season through the main competitive training period.

### 6.3. Methods

### 6.3.1. PARTICIPANTS

Twelve male ( 3 guards, 7 forwards, and 2 centers) and 11 female ( 3 guards, 6 forwards, and 2 centers) basketball players from the Portuguese Junior National Team volunteered to participate. All participants were elite junior players from the Portuguese National team and were aged 16 to 17 years old at the beginning of the season.

Participants inclusion criteria were: 1) > Tanner stage V (determined by selfevaluation [21]); 2) > 10 hours training per week; 3) negative test outcomes for performance-enhancing drugs; and 4) not taking any medications or dietary supplements. No females were taking oral contraceptives and were assessed during the luteal phase of their menstrual cycle, however two participants presented irregular cycles. Medical screening indicated that all participants were in good health, without endocrine abnormalities that would limit their participation in the study. All participants' tutors were informed about the possible risks of the investigation before giving written informed consent to participate. All procedures were approved by the Ethics Committee of the Faculty of Human Kinetics, Technical University of Lisbon, and were conducted in accordance with the declaration of Helsinki for human studies of the World Medical Association [22].

### 6.3.2. Experimental Design

This study used a longitudinal approach over a season ( $\sim 34$ weeks). The beginning of the pre-season (T0) testing was performed in the first week of the pre-
season training period (September) and the competitive-period assessment (T1) occurred at the end of the in-season period, two to three weeks before the main National competition corresponding to the final four of the National Championship in the years of age category under-18 (May), and two months before the under-18 years European Championships division-B category (males, last week of July, and females, first week of August). The definition of these season periods for assessments was made to understand the changes that occurred from the beginning of the pre-season, where athletes came from an 8 -week resting period, without any basketball practice, to the main National competition, where athletes are deemed to be at their best playing performance.

During the entire season players lived and trained at the National HighPerformance Centre. The participants were not taking exogenous anabolic androgenic steroids or other drugs or substances expected to affect body composition, physical performance, and hormonal balance during the season.

The male and female training regimens consisted of 5 sessions per week with a total of 120 minutes each, divided in technical-tactical training (including endurance running, ball exercise, sprint running, and training game). Once a week each player participated in one game integrated in the National Championship and one train game. Twice a week athletes performed resistance training for 60 minutes. In a first phase, males resistance training consisted of 2 sets of 10 exercises with $20 / 25$ maximal repetitions (RM) focused on the, development of neuro-muscular adaptations of the main muscular groups. During this 6 week period special attention was also given to footwork exercises, jumping and running abilities and ocular-manual coordination. The second phase lasting approximately 16 weeks aimed the muscular hypertrophy with 3 sets of 12-16 RM during the first 6-8 weeks, followed by 8-12 RM in the remaining 810 weeks. Again, special attention was given to footwork and running abilities as well as jumping skills. A third phase corresponded to another 6-8 weeks of power training consisting in 3 sets of 6-8 RM with slow velocities exercises to major muscle groups. The last phase, lasting 4 weeks, lied on power training with 3 sets of explosive exercises, 4-6 MR. For females the first phase was similar ( 6 weeks) and was followed by an 8 week period of 12-16 RM focusing muscular hypertrophy preceded by 8-12 RM the rest of the season.

### 6.3.3. BODY COMPOSITION MEASUREMENTS

Participants came to the laboratory in the morning, once at the beginning of the pre-season and again at the competitive training period, having fasted and refrained from exercise and alcohol or stimulant beverages for at least 10-12 hours. All measurements were carried out in the same morning.

## Anthropometric variables

Participants were weighed without shoes to the nearest 0.01 kg minimal clothes on an electronic scale connected to the plethysmograph computer (BOD POD ${ }^{\odot}$ COSMED, Rome, Italy). Based on 10 young active adults ( 5 males and 5 females), the coefficient of variation (CV) and technical error of measurement (TEM) for body mass in our laboratory were $0.07 \%$ and 0.04 kg , respectively. Height was measured to the nearest 0.1 cm with a stadiometer (Seca, Hamburg, Germany), according to the standardized procedures described elsewhere [23]. Based on 9 male elite athletes the CV and TEM for height were $0.04 \%$ and 0.06 cm , respectively. A certified anthropometrist performed the skinfold (SKF) measurements according to standardized procedures [23] using a Slim Guide caliper (Creative Health Products, Ann Arbor, Michigan, U.S.A.). Skinfold measurements included triceps, subscapular, biceps, suprailiac, abdominal, thigh, and medial calf, and the TEM, based on 24 elite athletes ( 13 males and 11 females), ranged from 0.20 to 0.27 mm . The sum of the 7 skinfolds ( $\Sigma 7 \mathrm{SKF}$ ) was used. Arm, thigh, and calf circumferences were measured according to standard procedures [23] and converted into muscle circumferences as circumference ( $\pi \mathrm{SKF}$ ) [24]. The circumferences were corrected for triceps, thigh, and calf SKF, respectively for arm, thigh and calf muscle circumferences. The TEM, based on 24 elite athletes ( 13 males and 11 females), were $0.08,0.15$, and 0.06 cm , respectively for arm, thigh, and calf circumferences.

## Hydration Status

The urine specific gravity (USG) was determined by a refractometer (Urisys 1100 Urine Analyzer, Roche, Portugal). The analyzer was calibrated with a control-Test (Chemstrip 10 MD ) every 7 days. After the dipsticks were inserted into the urine tubes they were placed and analyzed by the equipment, according to the manufacture
standardized procedures. Based on test-retest in 10 young active adults the CV and the ETM for the USG technique is $0.1 \%$ and 0.002 , respectively.

## Total Body Water

Total body water (TBW) was assessed by deuterium dilution using a stable Hydra gas isotope ratio mass spectrometer (PDZ, Europa Scientific, U.K.). After a 12hour fast, an initial urine sample was collected, followed by the administration of a deuterium oxide solution dose $\left({ }^{2} \mathrm{H}_{2} \mathrm{O}\right)$ of $0.1 \mathrm{~g} / \mathrm{kg}$ of body mass. After a 4-hour equilibration period, a second urine sample was collected. TBW was estimated, including a $4 \%$ correction due to TBW exchanging with non-aqueous compartments [25]. Based on test-retest using 10 elite male athletes, the CV and the TEM for TBW with the stable isotope ratio mass spectrometry in this laboratory were $0.3 \%$ and 0.11 kg , respectively.

## Extracellular Fluids

Extracellular water (ECW) was assessed by sodium bromide dilution. Subjects were asked to drink $0.030 \mathrm{~g} / \mathrm{kg}$ of body mass of NaBr . The NaBr concentration in plasma was measured by high-performance liquid chromatography (Dionex Corporation, Sunnyvale, California) before and 3 hours after tracer administration. The volume of ECW was calculated as: $\mathrm{ECW}(\mathrm{L})=$ [dose/(post-plasma bromide ([Br] PLASMA ) $-\operatorname{pre}([\mathrm{Br}]$ PLASMA $))] \times 0.90 \times 0.95$, where 0.90 is a correction factor for intracellular bromide $\left(\mathrm{Br}^{-}\right)$found mainly in red blood cells, and 0.95 is the Donnan equilibrium factor [25]. Extracellular fluids (ECF) were calculated as ECW $\times(1 / 0.98)$. Based on test-retest using 7 elite male athletes, the CV for ECW was $0.4 \%$, and the TEM was 0.08 kg .

## Intracellular Fluids

Intracellular fluids (ICF) were calculated as the difference between TBW and ECW using the dilution techniques mentioned above (deuterium and sodium bromide, respectively).

## Dual-Energy X-Ray Absorptiometry (DXA)

Bone mineral content (BMC), appendicular lean-soft tissue (ALST), and leansoft tissue (LST) were assessed using DXA equipment. The same technician positioned the participants and performed a total body scan Hologic Explorer-W, fan-beam densitometer, software QDR for Windows version 12.4 (Hologic, Waltham, Massachusetts, U.S.A.), according to the protocol described by the manufacturer. For athletes who were taller than the scan area we used a validated procedure that consists in the sum of a head and a trunk plus limbs scans [26]. Based on test-retest using 5 males and 5 females, the CV and the TEM in our laboratory for BMC, ALST, and LST were $1.3 \%(0.03 \mathrm{~kg}), 1.2 \%(0.24 \mathrm{~kg})$, and $0.8 \%(0.34 \mathrm{~kg})$, respectively.

Skeletal muscle was calculated as $[1.19 \times \operatorname{ALST}(\mathrm{kg})]-1.65$ [27] and BMC was converted to bone mineral (Mo) by multiplying it by 1.0436 [28].

## Body cell mass (BCM)

At the cellular level, BCM was calculated as [29]:
$B C M=L S T_{D X A}-(E C F+E C S)$

Where LST DXA is LST, ECF is extracellular fluids obtained, and ECS is extracellular solids calculated as $1.732 \times$ Mo.

## Body Volume

Body volume (BV) was assessed by air displacement plethysmography (ADP) (BOD $\mathrm{POD}^{\oplus}$ COSMED, Rome, Italy). After voiding their bladder, each subject was weighed to the nearest gram while wearing a swimsuit. The ADP device was calibrated according to the manufacturer's instructions. The effects of clothing and hair were accounted for by using a bathing suit and a swim cap. Finally, thoracic gas volume ( $\mathrm{T}_{\mathrm{GV}}$ ) was measured in the BOD POD ${ }^{\circledR}$ by using a technique common to standard pulmonary plethysmography called the "panting maneuver." While wearing a nose clip, the subjects breathed through a tube; after 2 to 3 normal breaths, the airway occluded for 3 seconds at mid-exhalation. During this time, the subject was instructed to gently puff against the occlusion by alternately contracting and relaxing the diaphragm. All measurements were conducted with software version 1.68. The CV and TEM for BV,
based on test-retest using 10 young active adults ( 5 males and 5 females), were $0.4 \%$ and 0.20 L , respectively.

## Four-component Model

A four-component model was used to assess body composition, calculated after using the total-body soft tissue mineral (Ms) component obtained as $\mathrm{Ms}=0.0129 \times$ TBW [30]. The model is described as follows:

$$
\begin{equation*}
F M(k g)=2.748 \times B V-0.699 \times T B W+1.129 \times M o-2.051 \times B M \tag{2}
\end{equation*}
$$

Where BV is body volume ( L ), TBW is total body water ( kg ), Mo is bone mineral ( kg ), and BM is body mass ( kg ).
The FFM was then calculated as BM minus FM.
Calculation of Density of Fat-free Mass ( $F F M_{D}$ ). The $\mathrm{FFM}_{\mathrm{D}}$ was estimated from TBW, Mo, Ms and protein (protein is equal to BM minus FM from the 4C model, TBW, Mo and Ms), contents of FFM (estimated as BM minus FM from the 4C model) and their densities ( $0.9937,2.982,3.317$, and $1.34 \mathrm{~g} / \mathrm{cm}^{3}$ ), for TBW, Mo, Ms and protein, respectively,
$\mathrm{FFM}_{\mathrm{D}}=1 /\left[\left(\mathrm{TBW} / \mathrm{TBW}_{\mathrm{D}}\right)+\left(\mathrm{Mo} / \mathrm{Mo}_{\mathrm{D}}\right)+\left(\mathrm{Ms} / \mathrm{MS}_{\mathrm{D}}\right)+\left(\right.\right.$ protein/protein $\left.\left.{ }_{\mathrm{D}}\right)\right]$

Where ${ }_{\mathrm{D}}$ is density, FFM is fat-free mass, TBW is total body water, Mo is bone mineral, and Ms is total-body soft tissue mineral.

Propagation Measurement Error. In the present study, the error associated with measurement of BCM and FM from the 4 C model, can be estimated by assuming an average body composition and measurement precision of each method. Accordingly,
$B C M \sigma^{2}=(1 \times 55.3 \times 0.008)^{2}+((1 / 0.98) \times 17.1 \times 0.004)^{2}+(1.732 \times 2.8 \times$ $0.013)^{2}=0.205(\sigma=0.45 \mathrm{~kg})$
$F M \sigma^{2}=(2.748 \times 64.5 \times 0.004)^{2}+(0.699 \times 43.6 \times 0.003)^{2}+(1.129 \times 2.8 \times$ $0.013)^{2}+(2.051 \times 71.4 \times 0.0007)^{2}=0.523(\sigma=0.72 \mathrm{~kg})$

Thus, the precision is 0.5 kg for BCM and 0.7 kg for FM .

### 6.3.4. PERFORMANCE TESTS

## Handgrip

Maximal isometric forearm strength (HGrip) was determined using a handgrip dynamometer (Jamar, Sammons Preston, Inc, Bolingbrook, IL, U.S.A.)) with visual feedback. The dynamometer was adjusted to each subject's dominant hand with each trial lasting approximately 5 -seconds. The best of three maximal trials was used for data analysis. The same adjustment of the dynamometer was used for all tests for each subject.

## Vertical Jump

Explosive power of the lower limbs was assessed by performing a countermovement jump abalakov (CMJ) in a custom contact platform (BioPlux System, version 1.0, Lisbon, Portugal). Participants were given detailed instructions and performed 2 trial jumps ( $\sim 50 \%$ of maximal height) with a resting period of 15 -seconds in between. The starting position was from upright standing position. They were then instructed to flex their knees $\left(90^{\circ}\right)$ as quickly as possible and then jump as high as possible with arm swing in the ensuing concentric phase. Subjects performed 3 jumps, with a 30 -seconds resting period and the jump with the greatest high was selected.

### 6.3.5. STATISTICAL ANALYSIS

Data analysis was performed using IBM SPSS Statistics version 19.0, 2010 (SPSS Inc., an IBM Company, Chicago, Illinois, U.S.A.). All analyses were performed separately for males and females. Descriptive statistics including means $\pm$ SD were calculated for all outcome measurements. Changes were expressed as a percentage of the baseline value. One sample t-tests were used to test changes that significantly differed from zero and to compare group means with the reference values based on cadaver analysis. Paired-samples t-tests were conducted to compare mean values of fatfree mass density $\left(\mathrm{FFM}_{\mathrm{D}}\right)$ and components between pre-season and competitive-period. Pearson correlations were used to analyse de association between body composition parameters and between body components with performance variables. Statistical significance was set at $\mathrm{p}<0.05$ (2-tailed).

Using a sample of 12 males and 11 females, prior data indicate that the difference in the response of matched pairs is approximately normally distributed with a standard deviation of nearly $3 \%$. Under such circumstances we have roughly $80 \%$ power to detect differences in the mean response of matched pairs of $-2.7 \%$ or $2.7 \%$ for males and of $-2.8 \%$ or $2.8 \%$ for females.

### 6.4. Results

Table 6.1. Body composition at the pre-season training period, competitive training period, and respective changes (results are expressed as mean $\pm$ SD).

|  | Male ( $\mathrm{n}=12$ ) <br> Age: $16.2 \pm 0.6$ (TO), $16.9 \pm 0.8$ (T1) |  |  | Females ( $\mathbf{n}=11$ ) <br> Age: $16.3 \pm 0.5$ (TO), $16.7 \pm 0.6$ (T1) |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | T0 | T1 | Changes** (\%) | то | T1 | Changes ${ }^{* *}$ (\%) |
| Whole body |  |  |  |  |  |  |
| Height (cm) | $192.5 \pm 6.5$ | $192.6 \pm 6.5$ | $0.08 \pm 0.15$ | $174.1 \pm 4.8$ | $174.8 \pm 4.4$ | $0.43 \pm 0.38^{+}$ |
| Body mass (kg) | $78.4 \pm 7.2$ | $80.9 \pm 7.6$ | $3.15 \pm 2.15^{+}$ | $63.7 \pm 6.9$ | $65.1 \pm 6.2$ | $2.37 \pm 2.46^{+}$ |
| BMI ( $\mathrm{kg} / \mathrm{m}^{2}$ ) | $21.2 \pm 1.8$ | $21.8 \pm 1.8$ | $2.98 \pm 2.08^{+}$ | $21.1 \pm 2.5$ | $21.3 \pm 2.1$ | $1.51 \pm 3.03$ |
| ¿7SKF (mm) | $69.1 \pm 16.9$ | $65.5 \pm 17.1$ | $-5.14 \pm 9.37$ | $106.1 \pm 35.2$ | $105.0 \pm 29.5$ | $0.57 \pm 13.99$ |
| Arm Mc (cm) | $25.7 \pm 1.9$ | $26.5 \pm 2.1$ | $3.40 \pm 2.73^{+}$ | $22.4 \pm 1.1$ | $23.2 \pm 1.1$ | $3.59 \pm 3.80^{+}$ |
| Thigh Mc (cm) | $48.3 \pm 3.3$ | $50.1 \pm 3.1$ | $3.81 \pm 2.95^{+}$ | $43.9 \pm 2.6$ | $45.5 \pm 2.0$ | $3.96 \pm 5.22^{+}$ |
| Calf Mc (cm) | $35.7 \pm 2.0$ | $35.9 \pm 2.0$ | $0.57 \pm 1.46$ | $31.2 \pm 2.3$ | $31.5 \pm 3.7$ | $0.80 \pm 10.10$ |
| Molecular |  |  |  |  |  |  |
| \%FM | $11.5 \pm 3.3$ | $10.4 \pm 3.8$ | $-8.56 \pm 22.86$ | $19.5 \pm 3.2$ | $20.7 \pm 3.7$ | $7.36 \pm 18.18$ |
| FM (Kg) | $9.0 \pm 2.8$ | $8.5 \pm 3.7$ | $-5.43 \pm 24.47$ | $12.5 \pm 2.9$ | $13.7 \pm 3.3$ | $10.01 \pm 19.30$ |
| FFM (kg) | $69.4 \pm 6.3$ | $72.4 \pm 6.0$ | $4.38 \pm 2.31{ }^{+}$ | $51.2 \pm 5.1$ | $51.5 \pm 3.7$ | $0.78 \pm 3.99$ |
| TBW (kg) | $49.5 \pm 6.0$ | $51.1 \pm 4.6$ | $3.51 \pm 4.56{ }^{\dagger}$ | $37.1 \pm 4.0$ | $35.9 \pm 2.3$ | $-2.69 \pm 6.5$ |
| Mo (kg) | $3.26 \pm 0.32$ | $3.43 \pm 0.35$ | $4.94 \pm 1.65{ }^{+}$ | $2.60 \pm 0.42$ | $2.69 \pm 0.44$ | $3.51 \pm 3.00^{+}$ |
| Protein (kg) | $15.9 \pm 1.7$ | $17.2 \pm 1.8$ | $8.02 \pm 7.60{ }^{+}$ | $11.0 \pm 1.2$ | $12.4 \pm 1.5$ | $13.00 \pm 12.06{ }^{\dagger}$ |
| Cellular |  |  |  |  |  |  |
| BCM (kg) | $38.5 \pm 3.3$ | $38.8 \pm 3.6$ | $0.99 \pm 3.10$ | $26.5 \pm 2.8$ | $26.6 \pm 2.1$ | $0.44 \pm 6.16$ |
| ECF (kg) | $19.7 \pm 1.5$ | $21.6 \pm 1.8$ | $9.36 \pm 6.00^{+}$ | $15.0 \pm 1.2$ | $16.4 \pm 1.3$ | $9.46 \pm 6.08^{+}$ |
| ICF (kg) | $30.2 \pm 4.7$ | $30.0 \pm 3.2$ | $0.12 \pm 8.83$ | $22.5 \pm 3.6$ | $19.9 \pm 1.3$ | $-9.67 \pm 13.60^{+}$ |
| Tissue-system |  |  |  |  |  |  |
| SM (kg) | $34.0 \pm 3.0$ | $35.5 \pm 3.2$ | $4.55 \pm 2.30^{+}$ | $22.4 \pm 2.2$ | $23.7 \pm 1.9$ | $5.90 \pm 4.63^{+}$ |
| ALST (kg)* | $29.9 \pm 2.5$ | $31.2 \pm 2.7$ | $4.34 \pm 2.19^{+}$ | $20.2 \pm 1.9$ | $21.3 \pm 1.6$ | $5.48 \pm 4.29^{+}$ |
| Arms $_{\text {LST }}(\mathrm{kg}$ ) | $7.2 \pm 0.6$ | $7.5 \pm 0.7$ | $4.52 \pm 5.16^{+}$ | $4.4 \pm 0.4$ | $4.8 \pm 0.3$ | $9.58 \pm 7.04{ }^{+}$ |
| Legs $_{\text {LST }}(\mathrm{kg}$ ) | $22.8 \pm 2.1$ | $23.7 \pm 2.1$ | $4.32 \pm 1.65{ }^{+}$ | $15.8 \pm 1.6$ | $16.4 \pm 1.4$ | $4.47 \pm 4.93^{+}$ |
| Performance |  |  |  |  |  |  |
| HGrip (kg) ${ }^{\ddagger}$ | $41.6 \pm 7.1$ | $47.6 \pm 4.9$ | $16.46 \pm 16.70$ | $30.2 \pm 4.0$ | $33.3 \pm 5.5$ | $9.62 \pm 6.16^{+}$ |
| CMJ (cm) ${ }^{\ddagger}$ | $35.6 \pm 4.4$ | $39.1 \pm 4.2$ | $10.16 \pm 9.42^{+}$ | $27.1 \pm 3.4$ | $29.9 \pm 5.0$ | $10.48 \pm 11.66^{\dagger}$ |

Abbreviations: T0, pre-season training period; T1, competitive training period; BMI, body mass index; $\sum 7$ SKF, sum of triceps, subscapular, biceps, supraspinale, abdominal, thigh, and medial calf skinfolds; Mc, muscle circumference; FM, fat mass; FFM, fat-free mass; TBW, total body water; Mo, bone mineral; BCM, body cell mass; ECF, extracellular fluids; ICF, intracellular fluids; SM, skeletal muscle; ALST, appendicular lean soft tissue; LST, lean soft tissue; HGrip, handgrip; CMJ, countermovement jump.
${ }^{* *}$ Changes are calculated as: T1 minus T0; ${ }^{\dagger}$ Changes significantly different from 0 ; *ALST was used to estimate SM;
${ }^{\ddagger}$ Data not available for 3 and 2 females for HGrip and CMJA, respectively.

In Table 6.1, body composition variables are described at the whole body, molecular, cellular, and tissue-system level at the beginning of the pre-season, at the competitive-training period, and the respective changes.

At the whole body level analysis we verified that, in both males and females, the centers were the tallest and heaviest, while guards were the shortest and lightest. At the competitive period, male guards were $185.8 \pm 4.3 \mathrm{~cm}$ and $73.7 \pm 4.5 \mathrm{~kg}$, forwards were $193.1 \pm 5.1 \mathrm{~cm}$ and $82.9 \pm 8.1 \mathrm{~kg}$, and centers were $201.0 \pm 0.0 \mathrm{~cm}$ and $84.5 \pm 1.5 \mathrm{~kg}$. Female guards were $172.2 \pm 1.6 \mathrm{~cm}$ and $60.6 \pm 8.5 \mathrm{~kg}$, forwards were $174.6 \pm 4.6 \mathrm{~cm}$ and $64.9 \pm 2.1 \mathrm{~kg}$, and centers were $179.8 \pm 3.9 \mathrm{~cm}$ and $72.6 \pm 6.9 \mathrm{~kg}$.

We further verified that both at T 0 and T 1 the muscle circumferences were highly associated ( $\mathrm{r} \geq 0.8, \mathrm{p}<0.001$ ) with the predicted muscle mass and ALST, and the $\sum 7$ SKF was highly associated with FM ( $\mathrm{r} \geq 0.8, \mathrm{p}<0.001$ ). Nevertheless we found a positive association between changes in the CMJ with changes in Calf Mc ( $\mathrm{r}=0.53$, 0.012 ) and changes in the $\sum 7 \mathrm{SKF}$ ( $\mathrm{r}=-0.54, \mathrm{p}=0.012$ ), these associations remained significant after adjusting for sex. We further verified that changes in the handgrip test were associated with changes in the $\sum 7$ SKF ( $\mathrm{r}=-0.48, \mathrm{p}=0.033$ ) (Table 6.2).

Considering the entire sample (Table 6.1) both males and females significantly increased their body masses, 2.5 kg ( $\mathrm{p}<0.001$ ) and 1.4 kg ( $\mathrm{p}=0.011$ ), respectively. In males, this gain was accompanied by an increase in FFM of $\sim 4 \%$ ( 3.0 kg ) ( $\mathrm{p}<0.001$ ) and all its main components (TBW, Mo, and protein). In females, only protein and Mo changed significantly from the pre-season to the competitive-training period. An increase of $\sim 5 \% ~(1.6 \mathrm{~kg} ; \mathrm{p}<0.001$ ) and $\sim 6 \% ~(1.3 \mathrm{~kg} ; \mathrm{p}=0.001$ ) in the SM in males and females was observed, respectively.

We further verified that changes in $\mathrm{FM}(\mathrm{r}=-0.53, \mathrm{p}=0.014)$, \%FM ( $\mathrm{r}=-0.54$, $\mathrm{p}=0.012$ ), and FFM ( $\mathrm{r}=0.46, \mathrm{p}=0.035$ ) were associated with changes in the CMJ test (Table 6.2), even adjusting for sex.

Table 6.2. Associations between body composition and performance at the pre-season training period, competitive training period, and respective changes.

|  | Hangrip Test (r) ${ }^{+}$ |  | CMJ test (r) ${ }^{\ddagger}$ |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | то | T1 | Changes** | то | T1 | Changes** |
| Whole body |  |  |  |  |  |  |
| Body mass | $0.74{ }^{*}$ | $0.76{ }^{*}$ | -0.53* | 0.46 * | 0.37 | -0.12 |
| BMI | 0.17 | $0.22{ }^{*}$ | -0.43 | -0.16 | -0.17 | -0.11 |
| S7SKF | -0.34 | -0.54* | -0.48* | -0.54* | -0.71* | -0.54* |
| Arm Mc | 0.62 * | $0.77 *$ | -0.20 | 0.50 * | $0.54 *$ | -0.20 |
| Thigh Mc | $0.68{ }^{*}$ | $0.72{ }^{*}$ | 0.09 | $0.45 *$ | $0.48{ }^{*}$ | -0.05 |
| Calf Mc | $0.64 *$ | $0.69 *$ | -0.14 | $0.53 *$ | $0.70{ }^{*}$ | $0.54 *$ |
| Molecular |  |  |  |  |  |  |
| FM | -0.12 | -0.44* | -0.36 | -0.51* | -0.70* | -0.53* |
| FFM | 0.75 * | $0.85 *$ | 0.07 | $0.60{ }^{*}$ | $0.59 *$ | 0.46 * |
| TBW | 0.75 * | $0.87{ }^{*}$ | 0.16 | $0.59 *$ | $0.61{ }^{*}$ | 0.33 |
| Cellular |  |  |  |  |  |  |
| BCM | $0.81 *$ | $0.84 *$ | -0.13 | $0.65 *$ | 0.60 * | 0.27 |
| ECF | 0.76 * | $0.83{ }^{*}$ | -0.22 | $0.62{ }^{*}$ | $0.52{ }^{*}$ | -0.15 |
| ICF | 0.71 * | $0.86{ }^{*}$ | 0.22 | $0.54 *$ | $0.64 *$ | 0.32 |
| Tissue-system |  |  |  |  |  |  |
| SM | $0.83 *$ | $0.85 *$ | -0.17 | $0.67{ }^{*}$ | $0.64 *$ | 0.22 |
| ALST | $0.83{ }^{*}$ | $0.85{ }^{*}$ | -0.17 | $0.67{ }^{*}$ | $0.64{ }^{*}$ | 0.22 |
| Arms $_{\text {LST }}$ | 0.80 * | $0.85{ }^{*}$ | -0.34 | $0.73{ }^{*}$ | $0.64{ }^{*}$ | 0.04 |
| Legs ${ }_{\text {LSt }}$ | $0.83 *$ | $0.84 *$ | 0.02 | $0.63{ }^{*}$ | 0.63* | 0.25 |

Abbreviations: T0, pre-season training period; T1, competitive training period; BMI, body mass index; $\Sigma 7 \mathrm{SKF}$, sum of triceps, subscapular, biceps, supraspinale, abdominal, thigh, and medial calf skinfolds; Mc, muscle circumference; FM, fat mass; FFM, fat-free mass; TBW, total body water; BCM, body cell mass; ECF, extracellular fluids; ICF, intracellular fluids; SM, skeletal muscle; ALST, appendicular lean soft tissue; LST, lean soft tissue; CMJ, countermovement jump.
*Significant correlations ( $\mathrm{p}<0.05$ ); **Changes are calculated as: T1 minus T0; ${ }^{\dagger}$ Data not available for 3 females (T0 and changes); ${ }^{\ddagger}$ Data not available for 2 females (T0 and changes) and 1 female (T1).

At the cellular level, athletes from both sexes, changed their ECF by $\sim 9 \%$ ( $\mathrm{p}<0.001$ ), whereas females reduced their ICF by $\sim 10 \%$ ( $\mathrm{p}=0.040$ ), which corresponded to a decrease of 2.6 kg in ICF. Figure 6.1 illustrates the differences in body composition at several levels of analysis.

*Significantly different from 0 ( $p<0.05$ )
Abbreviations: FM, fat mass; FFM, fat-free mass; TBW, total body water; ECF, extracellular fluids; ICF, intracellular fluids; BCM, body cell mass; SM, skeletal muscle
Figure 6.1. Differences in whole body, molecular, cellular, and tissue-system body composition levels of analysis.

At the molecular level, further analyses were performed (Figure 6.2) concerning $\mathrm{FFM}_{\mathrm{D}}$ and the contribution of its main components, namely, water, protein, and mineral [ $\mathrm{Mo}+\mathrm{Ms}$ ].

Concerning the reference values based on cadaver analysis, both males and females significantly differed from the established values for the water, protein, and mineral fraction. The $\mathrm{FFM}_{\mathrm{D}}$ values were not different from $1.1 \mathrm{~g} / \mathrm{cm}^{3}$, with the exception of the females' sample at the competitive period. Females significantly changed their protein and water FFM fractions and the $\mathrm{FFM}_{\mathrm{D}}$ from the pre-season to the competitive-training period.


Figure 6.2. Fat-free mass density and the contribution of water, protein, and mineral components at the beginning of the pre-season and at the end of the competitive period.

### 6.5. Discussion

The present study is the first to document body composition of elite junior basketball players at the beginning of a pre-season and at the main national competitive period using the five-level model approach for body composition analysis [13].

### 6.5.1. Molecular Level.

Numerous studies have assessed body composition at the molecular level in basketball players; however, most of them used field methods [10, 12, 31-33] to estimate molecular compartments and the majority refer to cross-sectional assessment. In our study, from the beginning of the pre-season to the main national competitive period, we observed an increase in body mass but no significant changes in FM in both sexes. At the molecular level, an increase in FFM was found in male players. We observed that athletes with a decrease in FM and an increase in FFM, improved vertical
jump, which is a commonly used test to assess the physical performance-related characteristics in basketball $[3,11,14,20]$.

Caterisano et al. [10] also assessed $\%$ FM at the pre- and post-season and did not observe any significant differences between assessments in male basketball players. In this study the density of games per week was similar to our sample, but the competitive season lasted 3 months, while in our investigation we assessed changes over an $\sim 8$ month period. The training regimen was similar to our players including resistance training 2 times per week. In the reported study, the sample had $\sim 6 \%$ FM, a value that is different from the one we observed in our sample ( $\sim 11 \%-10 \%$ ); however, these players were older ( $\sim 21$ years old) than our sample. Moreover like the majority of longitudinal studies in basketball, the authors used anthropometry to assess body composition. Tavino et al. [12] observed a decrease in \%FM from the beginning to the end of the preseason and an increase from the end of the pre-season to the competitive period using a sample of male basketball players aged 18 to 22 years old. In Tavino et al. [12] investigation, from the pre-season to the in-season, the players stopped performing weight training and reduced the training volume during the week. This training volume may justify the increase in $\% \mathrm{FM}$ observed from the end of the pre-season to the inseason. In our study, coaches focused on including weight training on players' regimens during the entire season and no changes were found in $\% \mathrm{FM}$ from the pre-season to the main competitive period. However, as only two assessments were performed, it might be speculated that our conclusions would be similar to these authors, as it is expected that body composition would improve during the pre-season and would be maintained or might decrease during the in-season [3]. On the other hand we aimed to verify the changes that occurred from the beginning of the pre-season, where athletes came from a resting period with lower fitness levels, to the main National competition, where athletes are deemed to be at their best playing performance.

Interestingly when looking at both cross-sectional and longitudinal studies regarding body composition, some did not mention or accurately describe body composition assessment [12, 32, 33]. Aforementioned studies evaluated elite basketball players, but if a coach wants to compare their athletes, the same method should be used to accurately compare each individual. In our study, we present information about the
methodology, providing coaches an accurate body composition profile they can use to compare their players.

Only few studies have reported cross-sectional FFM data of basketball players [16, 17], mostly based on 2C models to assess body composition. Athletes may have systematic deviations in $\mathrm{FFM}_{\mathrm{D}}$ from the value $1.1 \mathrm{~g} / \mathrm{cm}^{3}$ [34]. In our study, we observed that females' $\mathrm{FFM}_{\mathrm{D}}$ at the end of the competitive season differed from the established $1.1 \mathrm{~g} / \mathrm{cm}^{3}$ [35]. Variability in $\mathrm{FFM}_{\mathrm{D}}$ is one of the main factors limiting the accuracy of body composition estimates using densitometric models [36]. In our female participants, $\mathrm{FFM}_{\mathrm{D}}$ was significantly above $1.1 \mathrm{~g} / \mathrm{cm}^{3}$, and this may produce an underestimation of FM when using 2C models. In our sample, males presented FFM results at the competitive period similar to the results reported by Withers et al. [16, 17], though the athletes in Withers et al. [16, 17] investigation were considerably older ( $\sim 26$ years old). Likewise, in females the relative values of FFM at the competitive period are similar to Withers et al. study ${ }^{18,19}$ ( $\sim 79 \%$ of body mass), despite the fact that the players were older ( $\sim 23$ years old) and slightly heavier $(\sim 68 \mathrm{~kg})$ than the ones in our sample. In both studies [16, 17] FFM was assessed at the competitive period using underwater weighting, which estimates body composition based on densitometric models.

Portable bioimpedance analyzers have also been used to assess body composition in junior basketball players [31]; however, we demonstrated that, in our sample, FFM hydration deviated from the normal accepted $73.8 \%$ [35], and even considering the more consensual value of $73.2 \%$, based on studies with adult mammals [37], we still observed differences, with the exception of the results from females at the pre-season period. These findings suggest that, especially during a competitive period, where athletes' FFM hydration may be below the established values, methods that use hydrometric models as a principle may overestimate FM. Withers et al. [18] observed that most of the errors associated with 2C models lie not in the technical accuracy of the measurements, but in biological variability, which can be a serious threat to the validity of the assumed constants. To our knowledge, no study has characterized basketball players using a 4 C model. As we reported above, the most common methods used to assess body composition in these athletes are based on assumptions leading to possible errors, particularly if we want to track changes over a season.

### 6.5.2. Cellular Level

Cellular-level body composition analyses often are neglected in sports research; however, BCM and body fluid distribution are related to strength and aerobic performance $[38,39]$, in accordance we observed that both at the pre-season and at the competitive period BCM was associated with strength performance. Body cell mass can be defined as the total mass of "oxygen-exchanging, potassium-rich, glucose-oxidizing, work-performing" cells of the body [40]. To estimate BCM, we used an indirect model, and we did not observe any significant changes when comparing results from the preseason to the competitive period.

At this level a potential concern is the reduction observed in the intracellular fluids, as intracellular water reductions have been associated with a decrease in strength performance [8, 38], though in our study no association was observed. Being well hydrated is an important consideration for optimal exercise performance [5] and our cross-sectional results demonstrated a positive association between ICF and performance, given by the results in the handgrip and in the CMJ test.

### 6.5.3. Tissue-System Level

We observed a significant increase ( $\sim 6 \%$ ) in SM in the path of the season in both sexes. The emphasis given on weight training twice a week may have been important for the SM enhancing. Using magnetic resonance imaging (MRI) Midorikawa et al. [41], in a diverse sample of male athletes heavier ( $\sim 85 \mathrm{~kg}$ ) than ours ( $\sim 81 \mathrm{~kg}$ ), reported values of SM slightly below ( $\sim 33 \mathrm{~kg}$ ) the ones we observed in the competitive period ( $\sim 36 \mathrm{~kg}$ ). However, our study only comprises basketball players and Midorikawa et al. [41] sample consisted of athletes from different sports. Nevertheless, these differences may be related to the fact that we used an alternative method to estimate body composition. It is important to reinforce that MRI is the reference method for SM assessment. However this method is not available for the majority of laboratories, while the use of DXA to estimate ALST is an important predictor of SM that can be widely used to characterize the athletic population. A study [42] with a sample of adolescent athletes from several sports, slightly younger than our sample, reported values below the ones we observed at the competitive period, corresponding to $\sim 36 \%$ and $30 \%$ of
body mass, while in our results, ALST corresponded to $\sim 39 \%$ and $33 \%$ for males and females, respectively.

### 6.5.4. Whole Body Level

Although whole body methods are not considered the reference or the goldstandard techniques, often they are preferred by coaches to evaluate the effectiveness of the season planning training [20]. Nevertheless, in basketball, whole body methods might be important, as a player's body size largely determines his or her position on the court and also can be a determinant of success in junior elite basketball players [14]. Larger body mass is important in helping players maintain position when opponents challenge them under the basket [3]. In our sample, we observed that centers were the tallest and heaviest, while guards were the shortest and lightest, in accordance with other studies [31, 33, 43]. In our longitudinal data, we found a significant increase in body mass in both sexes; however, the majority of the studies have reported no significant differences in body mass in the course of a basketball season [10, 12, 15].

The sum of skinfolds is a practical technique to assess the body composition that has been included in the physiological assessment of basketball players [14, 15, 20]. In our sample, we used the $\sum 7$ SKF and, similarly to $\% \mathrm{FM}$, this variable did not significantly change between assessments, extending the findings reported by Hoffman et al. [15] that did not observe changes during the season in a sample of male basketball players ( $\sim 19$ years old). However, these authors used the sum of 8 skinfolds. Nevertheless we found that the $\sum 7$ SKF was highly correlated with FM assessed with the reference 4 C model reinforcing its usefulness in the field setting. At this regard, a negative association between changes in the $\sum 7$ SKF with changes in the handgrip and the CMJ test was verified, highlighting the importance of this practical variable on the field. Hoare [14], also using a sample of junior basketball players, observed that, in females, the $\sum 7$ SKF ranged from 83.5 to 108.7 mm , and, in males, it ranged from 57.5 to 70.0 mm , depending on the position. Also using a sample of junior basketball players, Stapff [20] reported mean values of 72.0 mm and 91.7 mm for males and females, respectively. In our sample, at the competitive period, males were slightly below the reported values ( 65.5 mm ), while females ( 105.0 mm ) seem to be considerably above. To our knowledge circumferences are not often used to assess body
composition in the athletic population, however it seems likely that arm, thigh, and calf muscle circumferences may be a useful tool as a representation of skeletal muscle [44]. Our data showed a positive association between changes in calf muscle circumference with changes in the CMJ test, demonstrating that players who increased their calf perimeter improved their vertical jump. Our results demonstrated a significant improvement in arm and thigh muscle circumferences, similarly to the results obtained in arms and legs LST and predicted SM. Moreover we found that all Mc were highly associated with SM. Anthropometric measures are easy and inexpensive and might be useful for coaches when assessing body composition changes during a season.

### 6.5.5. Study limitations

In our study we did not assess a control group to understand if the changes observed were effectively a result from the training season and not a function of growth development. However, our adolescents were at least in Tanner stage V [21] and Kim et al. [27], observed that an adult DXA SM prediction formula could be accurately applied to children and adolescents at a late stage of puberty (Tanner V), but not to prepubertal children or to children in earlier puberty (Tanner stage IV). Moreover, Molgaard and Michaelsen [45] conducted a one year body composition follow-up study in a healthy pediatric population aged 5-19 years using DXA. At the 16 to 17.9 age interval, the LST rate increase was $\sim 1.4 \mathrm{~kg} /$ year in boys and $\sim 0.1 \mathrm{~kg} /$ year in girls. Considering the 34 weeks of the season we would expect a LST increase of $0.9 \mathrm{~kg} /$ year and $0.07 \mathrm{~kg} /$ year due the effect of growth, respectively for males and females. Considering this values, and using one-sample T-test, we observed that in both males and females the increase of LST assessed by DXA in our sample ( $\sim 2.5 \mathrm{~kg}$ and $\sim 1.6 \mathrm{~kg}$, for males and females, respectively) was significantly different from the expected differences resulted from the growth process (data not shown), reinforcing the effect of the training season in body composition changes, even though a potential effect of growth may have occurred specifically on males. Nevertheless we address the absence of a control group as a limitation of this study.

### 6.6. Conclusions

We observed that a single basketball season was associated with significant differences at the molecular, cellular, tissue-system, and whole body level of body composition in both male and female junior players. Males increased their FFM, TBW, SM, and arm and calf Mc while the most important changes in females were the increase in SM and arm and calf Mc, and unexpectedly, the decrease in ICF. Both sexes improved their performance, specifically the handgrip and the vertical jump test and improvements in performance were associated with favourable changes molecular and whole body components. Moreover, our results reinforce the idea that densitometric and hydrometric methods in the athletic population must be used carefully, as assumed constants are the cornerstones of these methods.

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# Dual Energy X-Ray Absorptiometry and Anthropometry Reference Values for Athletes ${ }^{4}$ 

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# Dual Energy X-Ray Absorptiometry and Anthropometry Reference Values for Athletes 

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### 7.1. Abstract

Aim: Despite the importance of body composition in the athletic population with respect to both sports performance and health criteria, there is a dearth of reference data for sexes and sport-specific body composition and anthropometric measurements.

Methods: In this study 898 athletes ( 264 females, 634 males) were assessed for body weight and height, a total of 798 athletes ( 240 females and 558 males) were assessed for anthropometric variables, and 481 athletes ( 142 females and 339 males) were evaluated with dual-energy X-ray absorptiometry (DXA). A total of 21 different sports were represented. Reference percentiles $\left(5^{\text {th }}, 25^{\text {th }}, 50^{\text {th }}, 75^{\text {th }}\right.$, and $\left.95^{\text {th }}\right)$ were calculated for each measured value, stratified by sex and sport.

Results: DXA outcomes consist of total and regional (sub-total, trunk, and appendicular) bone mineral content and density, absolute and percent fat mass, fat-free mass, and lean soft tissue for athletes by sex and sport. Additionally we present results of body composition divided by height ${ }^{2}$ (weight, fat, fat-free mass, and appendicular lean soft tissue). Anthropometry outcomes included weight, height, sum of skinfolds (7 skinfolds, appendicular skinfolds, trunk skinfolds, arm skinfolds, and leg skinfolds), circumferences (Hip, arm, midthigh, calf, and abdominal circumferences), and muscle circumferences (arm, thigh, and calf muscle circumferences).

Conclusion: These reference percentiles should be a helpful tool for sports professionals, in both laboratory and field settings, for body composition assessment in athletes.

Key Words: body composition; athletes; reference values; DXA

### 7.2. Introduction

Body composition assessment in the athletic population may help to optimize competitive performance and monitor the success of training regimens and thus is of considerable interest to sports professionals [1-3]. It has been stated that in athletes an improved body composition is associated with enhancements in cardiorespiratory fitness $[4,5]$ and strength parameters [6-8].

Nevertheless it is recognized that body composition may also be related to health complications as medical problems may arise in athletes with very low body mass, extreme mass changes due to dehydration, or eating disorders [9].

Body composition can be organized according to a comprehensive model that consists of five levels of increasing complexity: I atomic; II molecular; III cellular; IV tissue-system; and V whole-body [10]. The majority of studies regarding the athletic population are focused mainly on estimation of molecular compartments and the description of whole-body parameters.

The whole-body level of body composition characterizes its body size and configuration, often described by anthropometric measures such as body weight, skinfolds, circumferences and body mass index (BMI) among others [11].

On the other hand, the molecular level consists of six main components: water, lipid, protein, carbohydrates, bone minerals, and soft tissue minerals. Several models ranging from two to six components can be created at this level of analysis [11]. Due to its good precision, availability, and low radiation dose, dual energy X-ray absorptiometry (DXA) is a convenient and useful tool for body composition assessment [12]. For athletes, DXA measurement presents an excellent alternative to reference methods due to its speed (fan-beam densitometers), but also because the measurement is minimally influenced by water fluctuation [1, 13, 14]. Furthermore DXA allows regional in addition to total body composition estimates, characterizing fat mass (FM) and dividing fat-free mass (FFM) into two components, lean soft tissue (LST) and bone mineral content (BMC) [11, 12, 15].

Reference values for DXA results were already developed for North Americans aged 8 to 85 years old using the NHANES dataset [16]. However, to our knowledge no
study has presented reference values for body composition in the athletic population within sports. Thus, the aim of the current study was to provide reference data for anthropometry and DXA outputs for the male and female athletic population from different sports during the in-season training period.

### 7.3. Methods

### 7.3.1. PARTICIPANTS

Using a cross-sectional design, a total of 898 athletes ( 264 females, 634 males) were assessed for body weight and height, 798 athletes ( 240 females and 558 males) were assessed for anthropometry variables and 481 athletes (142 females and 339 males) were evaluated with DXA during the in-season period. The sample covers athletes involved in a total of 21 sports. In Table 1, sample sizes and descriptive statistics for age are provided for the three general classifications of outcomes listed above, stratified by sex and sport.

Athletes involved in this study were subject to the following inclusion criteria: 1) at least in Tanner stage V (determined by self-evaluation [17]; 2) $\geq 10$ hours training per week; 3) negative test outcomes for performance-enhancing drugs; and 4) not taking any medications. Medical screening indicated that all subjects were in good health. All subjects and parents or guardians were informed about the possible risks of the investigation before giving written informed consent to participate. All procedures were approved by the Ethics Committee of the Faculty of Human Kinetics, Technical University of Lisbon, and were conducted in accordance with the declaration of Helsinki for human studies of the World Medical Association [18].

Table 7.1. Number of participants and respective age by sport and sex.

| Sport | Sex | Weight and Height |  |  | Skinfolds and Circumferences |  |  | DXA |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | n | Age <br> (range) | $\begin{aligned} & \text { Age } \\ & \text { (mean } \pm \text { SD) } \end{aligned}$ |  | Age <br> (range) | Age <br> (mean $\pm$ SD) | N | Age <br> (range) | Age <br> (mean $\pm$ SD) |
| Archery and Shooting | F | 4 | 25-45 | $33.5 \pm 8.5$ | 4 | 25-45 | $33.5 \pm 8.5$ | 0 | NA | NA |
|  | M | 9 | 16-50 | $30.9 \pm 13.1$ | 9 | 16-50 | $30.9 \pm 13.1$ | 0 | NA | NA |
| Athletics ${ }^{\text {a }}$ | F | 32 | 16-30 | $21.8 \pm 4.1$ | 25 | 17-30 | $22.0 \pm 3.9$ | 16 | 16-30 | $21.3 \pm 4.1$ |
|  | M | 30 | 17-31 | $21.6 \pm 3.5$ | 23 | 17-31 | $21.9 \pm 3.6$ | 11 | 17-26 | $20.1 \pm 3.0$ |
| Basketball | F | 43 | 16-34 | $17.3 \pm 2.7$ | 39 | 16-34 | $17.4 \pm 2.8$ | 34 | 16-19 | $16.9 \pm 0.8$ |
|  | M | 47 | 16-18 | $16.8 \pm 0.7$ | 46 | 16-18 | $16.8 \pm 0.7$ | 45 | 16-18 | $16.8 \pm 0.7$ |
| Fencing | F | 4 | 18-25 | $20.5 \pm 3.1$ | 4 | 18-25 | $20.5 \pm 3.1$ | 0 | NA | NA |
|  | M | 12 | 17-24 | $20.6 \pm 2.5$ | 12 | 17-24 | $20.6 \pm 2.5$ | 0 | NA | NA |
| Gymnastics | F | 18 | 16-23 | $18.3 \pm 2.4$ | 18 | 16-23 | $18.3 \pm 2.4$ | 12 | 16-19 | $17.1 \pm 1.1$ |
|  | M | 20 | 16-31 | $21.2 \pm 4.3$ | 20 | 16-31 | $21.2 \pm 4.3$ | 2 | 16-17 | $16.5 \pm 0.7$ |
| Handball | F | 4 | 19-31 | $25.3 \pm 4.9$ | 4 | 19-31 | $25.3 \pm 4.9$ | 4 | 19-31 | $25.3 \pm 4.9$ |
|  | M | 37 | 17-38 | $21.4 \pm 4.8$ | 20 | 17-21 | $19.1 \pm 1.1$ | 37 | 17-38 | $21.5 \pm 4.8$ |
| Hockey Rink | F | 0 | NA | NA | 0 | NA | NA | 0 | NA | NA |
|  | M | 49 | 16-36 | $20.5 \pm 5.4$ | 48 | 16-36 | $20.4 \pm 5.4$ | 2 | 17-25 | $21.0 \pm 5.7$ |
| Korfball | F | 9 | 18-30 | $21.2 \pm 3.6$ | 9 | 18-30 | $21.2 \pm 3.6$ | 0 | NA | NA |
|  | M | 11 | 16-31 | $22.7 \pm 5.3$ | 11 | 16-31 | $22.7 \pm 5.3$ | 0 | NA | NA |
| Modern Pentathlon | F | 9 | 16-23 | $18.6 \pm 2.6$ | 8 | 16-23 | $18.8 \pm 2.7$ | 2 | 17-17 | $17.0 \pm 0.0$ |
|  | M | 14 | 16-28 | $19.9 \pm 4.4$ | 14 | 16-28 | $19.9 \pm 4.4$ | 5 | 16-24 | $18.8 \pm 3.3$ |
| Motorsport | F | 0 | NA | NA | 0 | NA | NA | 0 | NA | NA |
|  | M | 7 | 17-33 | $26.0 \pm 6.6$ | 7 | 17-33 | $26.0 \pm 6.6$ | 0 | NA | NA |
| Other combat sports ${ }^{\text {b) }}$ | F | 15 | 16-24 | $18.5 \pm 2.4$ | 11 | 16-23 | $18.5 \pm 2.2$ | 4 | 17-24 | $18.8 \pm 3.5$ |
|  | M | 34 | 16-29 | $21.1 \pm 4.1$ | 29 | 16-29 | $21.6 \pm 4.2$ | 13 | 17-29 | $22.5 \pm 4.2$ |
| Rowing | F | 8 | 16-31 | $23.4 \pm 6.7$ | 8 | 16-31 | $23.4 \pm 6.7$ | 1 | 16-16 | $16.0 \pm$. |
|  | M | 27 | 16-32 | $21.1 \pm 4.5$ | 27 | 16-32 | $21.1 \pm 4.5$ | 6 | 16-17 | $16.8 \pm 0.4$ |
| Rugby | F | 0 | NA | NA | 0 | NA | NA | 0 | NA | NA |
|  | M | 62 | 16-33 | $20.4 \pm 4.0$ | 62 | 16-33 | $20.4 \pm 4.0$ | 39 | 16-28 | $18.2 \pm 2.1$ |
| Sailing | F | 7 | 16-27 | $20.6 \pm 4.2$ | 7 | 16-27 | $20.6 \pm 4.2$ | 0 | NA | NA |
|  | M | 38 | 16-40 | $25.0 \pm 7.6$ | 37 | 16-40 | $25.1 \pm 7.7$ | 4 | 19-35 | $26.0 \pm 7.1$ |
| Soccer | F | 22 | 16-37 | $22.5 \pm 5.7$ | 22 | 16-37 | $22.5 \pm 5.7$ | 0 | NA | NA |
|  | M | 42 | 17-36 | $19.7 \pm 4.1$ | 17 | 18-36 | $22.3 \pm 5.5$ | 28 | 17-19 | $18.0 \pm 0.8$ |
| Surf | F | 1 | 33-33 | $33.0 \pm$. | 1 | 33-33 | $33.0 \pm$. | 1 | 33-33 | $33.0 \pm$. |
|  | M | 1 | 31-31 | $31.0 \pm$. | 1 | 31-31 | $31.0 \pm$. | 0 | NA | NA |
| Swimming | F | 26 | 16-20 | $17.2 \pm 1.3$ | 26 | 16-20 | $17.2 \pm 1.3$ | 22 | 16-20 | $17.0 \pm 1.2$ |
|  | M | 44 | 16-30 | $19.6 \pm 3.4$ | 42 | 16-30 | $19.4 \pm 3.3$ | 36 | 16-30 | $19.1 \pm 3.4$ |
| Tennis | F | 11 | 16-24 | $18.0 \pm 2.7$ | 10 | 16-24 | $18.1 \pm 2.8$ | 5 | 16-24 | $19.0 \pm 3.7$ |
|  | M | 23 | 16-34 | $20.4 \pm 5.2$ | 19 | 16-34 | $19.8 \pm 5.4$ | 11 | 16-34 | $23.6 \pm 5.3$ |
| Triathlon | F | 11 | 16-27 | $21.0 \pm 3.5$ | 8 | 16-27 | $21.7 \pm 4.0$ | 10 | 16-26 | $20.4 \pm 3.1$ |
|  | M | 41 | 16-35 | $23.0 \pm 5.4$ | 33 | 16-35 | $23.1 \pm 5.5$ | 38 | 16-35 | $22.9 \pm 5.4$ |
| Volleyball | F | 16 | 18-36 | $25.9 \pm 5.9$ | 16 | 18-36 | $25.9 \pm 5.9$ | 16 | 18-36 | $25.9 \pm 5.9$ |
|  | M | 17 | 23-33 | $27.8 \pm 2.5$ | 17 | 23-33 | $27.8 \pm 2.5$ | 17 | 23-33 | $27.8 \pm 2.5$ |
| Wrestling and Judo ${ }^{\text {c) }}$ | F | 24 | 16-33 | $20.4 \pm 5.3$ | 21 | 16-33 | $19.7 \pm 5.2$ | 15 | 16-33 | $22.3 \pm 5.8$ |
|  | M | 69 | 16-45 | $21.0 \pm 5.0$ | 64 | 16-37 | $20.6 \pm 4.1$ | 45 | 16-45 | $21.8 \pm 5.1$ |

[^4]
### 7.3.2. BODY COMPOSITION MEASUREMENTS

Subjects came to the laboratory in a fasted state, and had refrained from exercise and alcohol or stimulant beverages for at least 3 hours.

## Anthropometric measurements

Anthropometric measures were performed by two certified anthropometrists. Body weight was measured without shoes to the nearest 0.01 kg and height was measured to the nearest 0.1 cm with a stadiometer (Seca, Hamburg, Germany), according to the standardized procedures described elsewhere [19]. Skinfold (SKF) measurements were made according to standardized procedures [19] using a Slim Guide caliper (Creative Health Products, Ann Arbor, Michigan, U.S.A.). Skinfold measurement included triceps, subscapular, biceps, suprailiac, abdominal, thigh, and medial calf. The sum of the 7 skinfolds ( $\sum 7 \mathrm{SKF}$ ); sum of appendicular (triceps, biceps, thigh, and medial calf) skinfolds ( $\sum_{\text {app }}$ SKF); sum of trunk (subscapular, suprailiac, and abdominal) skinfolds ( $\sum_{\text {trunk }}$ SKF); sum of arm (triceps and biceps) skinfolds ( $\sum_{\text {arm }}$ SKF); and sum of leg (thigh and medial calf) skinfolds ( $\sum_{\operatorname{leg}} S K F$ ) were used. Hip, arm, midthigh, calf [19] and abdominal [20] circumferences were measured according to standard procedures using an anthropometric tape (Lufkin W606PM, Apex Tool Group, Sparks, Maryland U.S.A.). Arm, thigh, and calf circumferences were converted into muscle circumferences as circumference - (Л SKF) [21]. The circumferences were corrected for triceps, thigh, and calf SKF, respectively for arm, thigh, and calf muscle circumferences.

The anthropometric measurements were conducted by two anthropometrists and the intra and inter coefficients of variation, calculated based on five highly active males, are presented in Table 7.2.

Table 7.2. Intra and inter coefficient of variations (CV) for skinfolds measured by the two anthropometrists

| Skinfolds | Inter measurers CV |  | Circumferences | Inter measurers CV |
| :--- | :--- | :--- | :--- | :--- |
| Subscapular | $13.16 \%$ | Waist | $2.11 \%$ |  |
| Abdominal | $10.29 \%$ | Hip | $0.90 \%$ |  |
| Suprailiac | $11.08 \%$ | Thigh | $1.02 \%$ |  |
| Thigh | $5.86 \%$ | Calf | $0.67 \%$ |  |
| Medial Calf | $3.15 \%$ | Arm | $1.31 \%$ |  |
| Triceps | $3.68 \%$ |  |  |  |
| Biceps | $10.54 \%$ |  |  |  |

Dual energy X-ray absorptiometry (DXA)
Athletes underwent a whole-body DXA scan according to the procedures recommended by the manufacturer on a Hologic Explorer-W, fan-beam densitometer (Hologic, Waltham, Massachusetts, USA). The equipment measures the attenuation of X-rays pulsed between 70 and 140 kV synchronously with the line frequency for each pixel of the scanned image. Following the protocol for DXA described by the manufacturer, a step phantom with six fields of acrylic and aluminium of varying thickness and known absorptive properties was scanned to serve as an external standard for the analysis of different tissue components. For athletes who were taller than the scan area we used a validated procedure that consists in the sum of a head and a trunk plus limbs scans [22]. The same technician positioned the participants, performed the scan and executed the analysis (software QDR for Windows version 12.4, Hologic, Waltham, Massachusetts, USA) according to the operator's manual using the standard analysis protocol.

The DXA measurements included whole-body measurements of bone mineral content (BMC, g), bone mineral density (BMD, g/cm ${ }^{2}$ ), absolute fat mass ( $\mathrm{FM}, \mathrm{kg}$ ), percent FM (\%FM), fat-free mass (FFM, kg), and lean soft tissue (LST, kg). With the exception of BMD, the remaining variables were also presented for pre-defined subregions, including trunk, appendicular (arms + legs) and subtotal (whole-body minus the head) regions. Additionally from these measures the following variables were calculated: fat mass index $(\mathrm{FMI})=\mathrm{FM} /$ height $^{2}\left(\mathrm{~kg} / \mathrm{m}^{2}\right)$, fat-free mass index $($ FFMI $)=$

FFM/height ${ }^{2}\left(\mathrm{~kg} / \mathrm{m}^{2}\right)$, and appendicular lean soft tissue index $($ ALSTI $)=$ ALST $/$ height ${ }^{2}$ $\left(\mathrm{kg} / \mathrm{m}^{2}\right)$.

The coefficients of variation in our laboratory were based on 10 young active adults (five males and five females) are presented in Table 7.3.

Table 7.3. Coefficients of variation in our laboratory for Dual-energy X-ray Absorptiometry measurements

|  | Whole-body | Sub-total | Appendicular | Trunk |
| :--- | :--- | :--- | :--- | :--- |
| BMC | $1.3 \%$ | $0.9 \%$ | $0.9 \%$ | $2.5 \%$ |
| BMD | $1.4 \%$ |  |  |  |
| Absolute FM | $1.7 \%$ | $1.8 \%$ | $2.8 \%$ | $4.3 \%$ |
| Percent FM | $1.6 \%$ | $1.7 \%$ | $2.1 \%$ | $3.6 \%$ |
| FFM | $0.8 \%$ | $0.6 \%$ | $1.6 \%$ | $1.2 \%$ |
| LST | $0.8 \%$ | $0.6 \%$ | $1.2 \%$ | $1.3 \%$ |

Abbreviations: BMC, bone mineral content; BMD, bone mineral density; FM, fat-mass; FFM, fat-free mass; LST, lean soft tissue

### 7.3.3. STATISTICAL ANALYSIS

Analyses were performed to complete four tasks: 1) Estimate the reference percentiles for each outcome, stratified by sex and sport; 2) Test whether or not the mean for each outcome differs by sex, stratified by sport; 3) Within each outcome, identify sports for which the mean value is different from the others (if any), stratified by sex; and 4) test for an association between anthropometric variables and DXA outcomes.

## Estimating reference percentiles

As the sample sizes are very low in many of the outcome/sex/sport combinations, the reference percentiles were estimated through a parametric, empirical Bayesian framework, allowing the sharing of information across sports to augment our inference whenever we have at least two athletes' values. Within a given sex and sport, the athletes' outcome values are assumed to follow a Normal (Gaussian) distribution that can be characterized through its mean and precision (inverse variance); if the mean and precision are known, all quantiles follow immediately from the Normal assumption.

The sport-specific means and variances are modelled as arising from a NormalGamma, which serves as the prior and forms a conjugate family with our observational
model. The hyperparameters of the prior are informed empirically through maximumlikelihood using all athletes' data for this outcome, restricted by sex. Once this is done, joint posterior distributions for the mean and precision are generated for every sport, giving rise to point estimates and $95 \%$ joint confidence regions for the mean and precision, which in turn are used to calculate simultaneous $95 \%$ confidence intervals for the reference percentiles of interest.

All computation was performed in R version 2.14 .2 [23]. In some cases outcome values were decidedly non-Normal: BMD, FM, \%FM, FMI, FFMI, and ALSTI were logged before running the approach for all subjects and then transformed back afterwards in order to ameliorate this concern while maintaining the original units for all results.

## Comparisons across sex and sports

Descriptive statistics (mean and standard deviation) for the main outcome variables were calculated with IBM SPSS Statistics version 21.0, 2012 (IBM, Chicago, Illinois, U.S.A.). Normality was tested using the Shapiro-Wilk test. Sex comparisons were performed with unpaired T-tests or the non-parametric equivalent, the MannWhitney U test. Comparisons across sports were made by sex using the Kruskall-Wallis test with pairwise comparisons performed using the Dunn test. For both sex and sport comparisons the p-values presented are nominal, (i.e., unadjusted for multiple comparisons across outcomes and either sexes or sports, as appropriate).

## Association between anthropometric variables and DXA

Simple linear regression analysis was performed to verify if the $\sum 7$ SKF was associated with \%FM from DXA. Also, multiple linear regression analysis was performed to test if muscle circumferences (arm, thigh, and calf muscle circumferences) were associated with ALST. Normality, homogeneity, and homoscedasticity of the residuals were analysed. Regression analyses were performed with calculated with IBM SPSS Statistics version 21.0, 2012 (IBM, Chicago, Illinois, U.S.A.).

### 7.4. Results

The variables derived from DXA and anthropometry measures are provided in Table 7.4 Reference values for each of the DXA and anthropometry outputs described in Table 7.4 are provided as supplementary material (Table S1 to Table S40) by sport and sex. The percentiles include the $5^{\text {th }}, 25^{\text {th }}, 50^{\text {th }}, 75^{\text {th }}$, and $95^{\text {th }}$ percentile.

Table 7.4. List of percentiles derived from dual energy X-ray absorptiometry (DXA) and anthropometry measures.

| Anthropometry variable | Supplemental <br> Table | DXA variable | Supplementary <br> Table |
| :---: | :---: | :---: | :---: |
| Weight (kg) | S1 | WB BMC (g) | S17 |
| Height (cm) | S2 | WB BMD ( $\mathrm{g} / \mathrm{cm}^{2}$ ) | S18 |
| BMI (kg/m ${ }^{2}$ ) | S3 | WB FM (kg) | S19 |
| \7SKF (mm) | S4 | WB FMI ( $\mathrm{kg} / \mathrm{m}^{2}$ ) | S20 |
| SAppendicular SKF (mm) | S5 | WB FM (\%) | S21 |
| EArm SKF (mm) | S6 | WB FFM (kg) | S22 |
| $\sum$ Leg SKF (mm) | S7 | WB FFMI ( $\mathrm{kg} / \mathrm{m}^{2}$ ) | S23 |
| ETrunk SKF (mm) | S8 | WB LST (kg) | S24 |
| Arm circumference (cm) | S9 | Subtot BMC (kg) | S25 |
| Arm muscle circumference (cm) | S10 | Subtot FM (kg) | S26 |
| Thigh circumference (cm) | S11 | SubTot FM (\%) | S27 |
| Thigh muscle circumference (cm) | S12 | Subtot FFM (kg) | S28 |
| Calf circumference (cm) | S13 | Subtot LST (kg) | S29 |
| Calf muscle circumference (cm) | S14 | Appendicular BMC (kg) | S30 |
| Abdominal circumference (cm) | S15 | Appendicular FM (kg) | S31 |
| Hip circumference (cm) | S16 | Appendicular FM (\%) | S32 |
|  |  | Appendicular FFM (kg) | S33 |
|  |  | Appendicular LST (kg) | S34 |
|  |  | Appendicular LSTI (kg/m ${ }^{2}$ ) | S35 |
|  |  | Trunk BMC (kg) | S36 |
|  |  | Trunk FM (kg) | S37 |
|  |  | Trunk FM (\%) | S38 |
|  |  | Trunk FFM (kg) | S39 |
|  |  | Trunk LST (kg) | S40 |

Descriptive statistics for the main anthropometry and DXA variables are presented in Table 7.5 and 7.6, respectively.
Table 7.5. Body composition for the main anthropometry outputs.

| Sport | Gender | n | Weight (kg) ${ }^{\text {b.c }}$ | Height (cm) ${ }^{\text {b.c }}$ | BMI ( $\left.\mathrm{kg} / \mathrm{m}^{2}\right)^{\text {b.c }}$ | n | $\Sigma 7$ SKF (mm) ${ }^{\text {b.c }}$ | Arm Mc* ${ }^{(c m)}{ }^{\text {b.c }}$ | Thigh Mc* ${ }^{(c m)}{ }^{\text {b.c }}$ | Calf Mc* $(\mathrm{cm})^{\mathrm{b} . \mathrm{c}}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1. Archery and Shooting | F | $4^{\#}$ | $58.3 \pm 3.3^{\text {a }}$ | $162.7 \pm 1.4^{\text {a }}$ | $22.0 \pm 1.1$ | $4{ }^{\text {\# }}$ | $143.9 \pm 21.6^{\text {a }}$ | $20.5 \pm 1.4^{\text {a }}$ | $40.6 \pm 3.9^{\text {a }}$ | $28.5 \pm 1.3^{\text {a }}$ |
|  | M | 9 | $73.1 \pm 10.0$ | $177.6 \pm 4.2$ | $23.2 \pm 3.6$ | 9 | $91.6 \pm 29.2$ | $26.0 \pm 2.5$ | $44.3 \pm 2.1$ | $33.0 \pm 1.7$ |
| 2. Athletics | F | 32 | $59.1 \pm 6.3^{\text {a }}$ | $166.3 \pm 6.3^{\text {a }}$ | $21.3 \pm 1.4^{\text {a }}$ | 25 | $71.4 \pm 24.9^{\text {a }}$ | $22.6 \pm 1.6^{\text {a }}$ | $46.4 \pm 3.4^{\text {a }}$ | $33.0 \pm 1.9^{\text {a }}$ |
|  | M | 30 | $73.9 \pm 6.4$ | $182.1 \pm 5.5$ | $22.3 \pm 1.5$ | 23 | $46.6 \pm 11.6$ | $27.1 \pm 1.9$ | $51.2 \pm 3.7$ | $35.9 \pm 2.1$ |
| 3. Basketball | F | 43 | $68.9 \pm 9.4^{\text {a }}$ | $176.7 \pm 8.2^{\text {a }}$ | $22.0 \pm 2.0$ | 39 | $126.8 \pm 36.9^{\text {a }}$ | $21.8 \pm 1.7^{\text {a }}$ | $45.9 \pm 3.1^{\text {a }}$ | $31.8 \pm 2.7^{\text {a }}$ |
|  | M | 47 | $81.9 \pm 10.5$ | $190.6 \pm 9.9$ | $22.5 \pm 1.9$ | 46 | $73.3 \pm 23.6$ | $27.1 \pm 2.2$ | $50.8 \pm 3.3$ | $36.3 \pm 2.7$ |
| 4. Fencing | F | $4^{\text {\# }}$ | $61.7 \pm 4.9^{\text {a }}$ | $166.3 \pm 6.2^{\text {a }}$ | $22.3 \pm 2.1$ | $4^{*}$ | $121.5 \pm 17.0^{\text {a }}$ | $20.7 \pm 1.4^{\text {a }}$ | $45.3 \pm 2.5^{\text {a }}$ | $30.4 \pm 0.8^{\text {a }}$ |
|  | M | 12 | $72.6 \pm 7.2$ | $180.0 \pm 5.7$ | $22.4 \pm 1.7$ | 12 | $63.8 \pm 21.9$ | $25.0 \pm 1.3$ | $52.3 \pm 3.0$ | $34.9 \pm 2.2$ |
| 5. Gymnastics | F | 18 | $53.2 \pm 5.6^{\text {a }}$ | $160.7 \pm 6.8^{\text {a }}$ | $20.6 \pm 1.6^{\text {a }}$ | 18 | $91.4 \pm 23.3^{\text {a }}$ | $21.1 \pm 1.8^{\text {a }}$ | $45.4 \pm 4.0^{\text {a }}$ | $29.7 \pm 2.0^{\text {a }}$ |
|  | M | 20 | $65.9 \pm 6.8$ | $169.9 \pm 6.0$ | $22.9 \pm 2.7$ | 20 | $58.5 \pm 20.2$ | $26.8 \pm 2.0$ | $45.2 \pm 3.4$ | $33.4 \pm 2.5$ |
| 6. Handball | F | $4^{\text {\# }}$ | $67.9 \pm 4.9^{\text {a }}$ | $167.3 \pm 3.6^{\text {a }}$ | $24.2 \pm 1.6$ | $4^{\text {\# }}$ | $128.1 \pm 31.1^{\text {a }}$ | $24.2 \pm 1.8^{\text {a }}$ | $49.2 \pm 2.8^{\text {a }}$ | $32.5 \pm 1.7^{\text {a }}$ |
|  | M | 37 | $83.7 \pm 10.9$ | $183.4 \pm 6.2$ | $24.8 \pm 2.6$ | 20 | $86.7 \pm 31.3$ | $27.7 \pm 2.3$ | $48.9 \pm 3.5$ | $35.4 \pm 2.1$ |
| 7. Hockey Rink | F | $0^{*}$ |  |  |  | $0^{*}$ |  |  |  |  |
|  | M | 49 | $74.9 \pm 8.0$ | $174.8 \pm 5.1$ | $24.5 \pm 2.3$ | 48 | $80.7 \pm 28.0$ | $26.4 \pm 2.2$ | $49.2 \pm 3.0$ | $34.9 \pm 2.9$ |
| 8. Korfball | F | 9 | $58.5 \pm 6.0^{\text {a }}$ | $162.4 \pm 8.7^{\text {a }}$ | $22.2 \pm 1.9$ | 9 | $114.5 \pm 31.7^{\text {a }}$ | $20.9 \pm 0.7^{\text {a }}$ | $40.0 \pm 2.6{ }^{\text {a }}$ | $30.7 \pm 1.8^{\text {a }}$ |
|  | M | 11 | $72.3 \pm 7.8$ | $181.2 \pm 6.2$ | $22.0 \pm 2.1$ | 11 | $61.7 \pm 18.1$ | $25.3 \pm 2.6$ | $45.4 \pm 2.2$ | $35.7 \pm 1.6$ |
| 9. Modern Pentathlon | F | 9 | $60.8 \pm 9.4{ }^{\text {a }}$ | $170.6 \pm 6.5^{\text {a }}$ | $20.8 \pm 2.7$ | 8 | $87.0 \pm 27.4^{\text {a }}$ | $21.8 \pm 2.1^{\text {a }}$ | $44.7 \pm 1.5^{\text {a }}$ | $30.6 \pm 5.0^{\text {a }}$ |
|  | M | 14 | $69.0 \pm 8.2$ | $176.9 \pm 5.3$ | $22.1 \pm 2.5$ | 14 | $56.3 \pm 18.0$ | $26.7 \pm 2.2$ | $50.5 \pm 7.6$ | $34.5 \pm 2.0$ |
| 10. Motorsport | F | $0^{*}$ |  |  |  | $0^{*}$ |  |  |  |  |
|  | M | $7{ }^{\text {\# }}$ | $73.9 \pm 7.2$ | $175.5 \pm 5.0^{\text {a }}$ | $23.9 \pm 1.4$ | $7{ }^{\text {\# }}$ | $81.9 \pm 34.4$ | $26.3 \pm 2.5$ | $49.3 \pm 3.2$ | $34.0 \pm 1.9$ |
| 11. Other combat sports | F | 15 | $59.0 \pm 6.3$ | $162.8 \pm 6.4$ | $22.3 \pm 2.2$ | 11 | $107.3 \pm 39.4{ }^{\text {a }}$ | $22.6 \pm 1.7^{\text {a }}$ | $44.7 \pm 2.7^{\text {a }}$ | $31.6 \pm 1.4^{\text {a }}$ |
|  | M | 34 | $70.3 \pm 8.5$ | $175.9 \pm 6.1$ | $22.7 \pm 2.3$ | 29 | $62.6 \pm 20.4$ | $26.9 \pm 1.8$ | $49.9 \pm 2.8$ | $34.4 \pm 4.3$ |
| 12. Rowing | F | 8 | $66.1 \pm 6.2^{\text {a }}$ | $169.9 \pm 5.6^{\text {a }}$ | $22.9 \pm 1.3$ | 8 | $104.5 \pm 20.9^{\text {a }}$ | $22.1 \pm 1.3^{\text {a }}$ | $47.2 \pm 2.8^{\text {a }}$ | $31.3 \pm 1.2^{\text {a }}$ |
|  | M | 27 | $78.5 \pm 8.9$ | $183.0 \pm 5.6$ | $23.4 \pm 2.0$ | 27 | $60.5 \pm 19.3$ | $27.8 \pm 1.5$ | $50.1 \pm 2.7$ | $34.6 \pm 2.8$ |
| 13. Rugby | F | $0^{\# \#}$ |  |  |  | $0^{*}$ |  |  |  |  |
|  | M | 62 | $92.2 \pm 16.1$ | $182.8 \pm 7.5$ | $27.6 \pm 4.5$ | 62 | $110.5 \pm 57.2$ | $31.1 \pm 3.4$ | $52.5 \pm 4.7$ | $36.8 \pm 2.9$ |
| 14. Sailing | F | $7{ }^{\text {\# }}$ | $62.5 \pm 9.1^{\text {a }}$ | $170.2 \pm 9.6^{\text {a }}$ | $21.5 \pm 1.7^{\text {a }}$ | $7{ }^{\text {\# }}$ | $90.9 \pm 40.3$ | $23.2 \pm 3.3^{\text {a }}$ | $43.9 \pm 3.3^{\text {a }}$ | $32.4 \pm 6.5$ |
|  | M | 38 | $76.1 \pm 12.3$ | $177.9 \pm 7.7$ | $23.9 \pm 2.7$ | 37 | $89.7 \pm 35.4$ | $26.7 \pm 3.3$ | $48.1 \pm 6.2$ | $33.9 \pm 3.0$ |

Table 7.5 (cont.) Body composition for the main anthropometry outputs.

| Sport | Gender | n | Weight (kg) ${ }^{\text {b.c }}$ | Height (cm) ${ }^{\text {b.c }}$ | BMI ( $\left.\mathrm{kg} / \mathrm{m}^{2}\right)^{\text {b.c }}$ | n | V7SKF (mm) ${ }^{\text {b.c }}$ | Arm MC ${ }^{*}(\mathrm{~cm})^{\text {b.c }}$ | Thigh MC ${ }^{*}(\mathrm{~cm})^{\text {b.c }}$ | Calf MC ${ }^{*}(\mathrm{~cm})^{\text {b.c }}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 15. Soccer | F | 22 | $59.7 \pm 6.5^{\text {a }}$ | $164.1 \pm 5.7^{\text {a }}$ | $22.2 \pm 2.0$ | 22 | $105.5 \pm 26.8^{\text {a }}$ | $21.6 \pm 1.9^{\text {a }}$ | $44.7 \pm 3.5^{\text {a }}$ | $30.7 \pm 2.7^{\text {a }}$ |
|  | M | 42 | $73.8 \pm 7.7$ | $176.6 \pm 5.7$ | $23.6 \pm 1.9$ | 17 | $58.1 \pm 11.9$ | $27.7 \pm 2.3$ | $50.9 \pm 3.0$ | $35.2 \pm 2.9$ |
| 16. Surf | F | $1{ }^{\text {\# }}$ | 47.2 | 149.7 | 21.1 | $1{ }^{\text {\# }}$ | 72.0 | 21.9 | 42.3 | $30.8{ }^{\text {a }}$ |
|  | M | $1{ }^{\text {\# }}$ | 71.2 | 167.0 | 25.5 | $1{ }^{\text {\# }}$ | 78.5 | 26.9 | 44.4 | 32.7 |
| 17. Swimming | F | 26 | $59.3 \pm 5.4^{\text {a }}$ | $167.7 \pm 5.9^{\text {a }}$ | $21.1 \pm 1.5^{\text {a }}$ | 26 | $93.1 \pm 26.0^{\text {a }}$ | $23.9 \pm 1.9^{\text {a }}$ | $44.2 \pm 2.7^{\text {a }}$ | $31.2 \pm 1.9^{\text {a }}$ |
|  | M | 44 | $72.0 \pm 8.8$ | $179.9 \pm 7.0$ | $22.2 \pm 1.8$ | 42 | $56.6 \pm 19.8$ | $30.9 \pm 13.7$ | $48.2 \pm 2.9$ | $34.4 \pm 1.8$ |
| 18. Tennis | F | 11 | $64.2 \pm 6.4^{\text {a }}$ | $168.5 \pm 5.2^{\text {a }}$ | $22.6 \pm 1.6$ | 10 | $141.0 \pm 24.7^{\text {a }}$ | $21.9 \pm 3.4^{\text {a }}$ | $41.0 \pm 4.2^{\text {a }}$ | $29.8 \pm 1.9^{\text {a }}$ |
|  | M | 23 | $71.3 \pm 9.7$ | $177.4 \pm 6.1$ | $22.6 \pm 2.6$ | 19 | $67.7 \pm 17.4$ | $25.6 \pm 2.3$ | $46.7 \pm 3.5$ | $34.1 \pm 1.8$ |
| 19. Triathlon | F | 11 | $57.9 \pm 6.5^{\text {a }}$ | $168.4 \pm 6.8^{\text {a }}$ | $20.4 \pm 2.0$ | $8{ }^{\text {\# }}$ | $86.3 \pm 43.4$ | $23.2 \pm 2.6^{\text {a }}$ | $45.3 \pm 3.4$ | $32.5 \pm 2.2^{\text {a }}$ |
|  | M | 41 | $65.9 \pm 4.2$ | $175.8 \pm 5.6$ | $21.3 \pm 1.3$ | 33 | $49.6 \pm 11.3$ | $26.2 \pm 1.5$ | $47.1 \pm 2.5$ | $34.2 \pm 1.5$ |
| 20. Volleyball | F | 16 | $67.7 \pm 11.4{ }^{\text {a }}$ | $174.5 \pm 9.9^{\text {a }}$ | $22.1 \pm 2.0^{\text {a }}$ | 16 | $118.0 \pm 35.3^{\text {a }}$ | $23.0 \pm 2.4^{\text {a }}$ | $46.5 \pm 3.8^{\text {a }}$ | $32.5 \pm 2.5^{\text {a }}$ |
|  | M | 17 | $90.1 \pm 7.4$ | $195.0 \pm 6.7$ | $23.7 \pm 1.6$ | 17 | $70.0 \pm 19.7$ | $31.1 \pm 2.9$ | $52.5 \pm 2.7$ | $36.9 \pm 2.1$ |
| 21. Wrestling and Judo | F | 24 | $59.4 \pm 9.0^{\text {a }}$ | $162.1 \pm 6.8^{\text {a }}$ | $22.5 \pm 2.2^{\text {a }}$ | 21 | $123.0 \pm 41.7^{\text {a }}$ | $22.3 \pm 2.7^{\text {a }}$ | $44.3 \pm 3.8^{\text {a }}$ | $29.1 \pm 3.2^{\text {a }}$ |
|  | M | 69 | $71.5 \pm 9.4$ | $172.7 \pm 6.4$ | $23.9 \pm 2.5$ | 64 | $59.4 \pm 23.1$ | $29.0 \pm 3.2$ | $49.4 \pm 2.7$ | $34.4 \pm 2.8$ |
| All sample | F | 264 | $61.5 \pm 8.7^{\text {a }}$ | $167.8 \pm 8.6^{\text {a }}$ | $21.8 \pm 2.0^{\text {a }}$ | 240 | $106.8 \pm 36.5^{\text {a }}$ | $22.3 \pm 2.2^{\text {a }}$ | $44.9 \pm 3.6^{\text {a }}$ | $31.2 \pm 2.8^{\text {a }}$ |
|  | M | 634 | $76.1 \pm 12.2$ | $179.3 \pm 8.6$ | $23.6 \pm 3.0$ | 558 | $71.4 \pm 33.8$ | $27.9 \pm 4.8$ | $49.5 \pm 4.2$ | $35.0 \pm 2.8$ |
| Sport Comparisons | F |  | $\begin{aligned} & { }^{\text {b) }} 5 \neq 18,12,20,3 ; \\ & 2,21 \neq 3 \end{aligned}$ | $\begin{aligned} & { }^{\text {b) }} 5,21,11 \neq 20,3 ; \\ & 8,15 \neq 3 \end{aligned}$ | NS |  | $\begin{aligned} & { }^{\text {b) }} 2 \neq 15,20,21,3, \\ & 18,1 ; 17 \neq 3,18 ; 5 \\ & \neq 18 \end{aligned}$ | ${ }^{\text {b) }} 8,5,15 \neq 17$ | $\begin{aligned} & \text { b) } 8 \neq 3,2,20,12,6 ; \\ & 18 \neq 2,6 \end{aligned}$ | ${ }^{\text {b) }} 21 \neq 20,2,3 ; 5 \neq 2$ |
|  | M |  | $\begin{aligned} & { }^{\text {c) }} 5,19, \neq 7,12,3 \\ & 6,20,13 ; 19 \neq 15 \\ & 2,14 ; 9,18,11, \neq \\ & 3,6,20,13 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 5,21 \neq 14,17,2, \\ & 13,12,6,3,20 ; 5 \neq \\ & 8 ; 7 \neq 2,13,12,6, \\ & 3,20 ; 10 \neq 3,20 ; \\ & 19 \neq 2 \end{aligned}$ | $\begin{array}{r} { }^{\text {c) }} 19,2 \neq 3,1,14,7 \\ 6,13 ; 2 \neq 20 ; 9 \neq \\ 13 ; 17,21 \neq 14,7, \\ 6,13 \end{array}$ |  | $\begin{aligned} & { }^{\text {c) }} 2,19 \neq 3,7,6,14, \\ & 1,13 ; 2 \neq 20 ; 9 \neq \\ & 13 ; 17,21 \neq 7,6 \\ & 14,13 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 4 \neq 17 ; 4,8,18, \\ & 19, \neq 21,20,13 ; 1 \\ & \neq 20,13 \end{aligned}$ | $\begin{aligned} & { }^{c} \text { ) } 1,5 \neq 21,11 ; 5 \neq 7 \\ & 1,5,8,19, \neq 12,3 \\ & 15,2,4,20,13 ; 18 \neq \\ & 3 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 1,5 \neq 3,13,20 ; 18 \\ & \neq 13 \end{aligned}$ |

[^5]Table 7.6. Body composition for the Dual-energy X-ray absorptiometry outputs.

| Sport | gender | n | $\underset{\mathrm{b}, \mathrm{c}}{\mathrm{WB}} \text { BMC (g) }$ | $\begin{aligned} & \text { WB BMD } \\ & \left(\mathrm{g} / \mathrm{cm}^{2}\right)^{\mathrm{b}, \mathrm{c}} \end{aligned}$ | $\begin{aligned} & \text { WB FM } \\ & (\mathrm{kg})^{\mathrm{b}, \mathrm{c}} \end{aligned}$ | $\underset{b, c}{\text { WB FM (\%) }}$ | $\underset{b, c}{\mathrm{FMII}}\left(\mathrm{~kg} / \mathrm{m}^{2}\right)$ | $\begin{aligned} & \text { WB FFM } \\ & {\mathbf{( k g})^{b, c}}^{2} \end{aligned}$ | $\begin{aligned} & \text { FFMI } \\ & \left(\mathbf{k g} / \mathbf{m}^{2}\right)^{b, c} \end{aligned}$ | $\underset{\mathrm{b}, \mathrm{c}}{\mathrm{~W}} \text { LST (kg) }$ | ALST (kg) ${ }^{\text {b, c }}$ | $\begin{aligned} & \text { ALSTI } \\ & \left(\mathbf{k g} / \mathrm{m}^{2}\right)^{\mathrm{b}, \mathrm{c}} \end{aligned}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1. Archery+Shooting | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
| 2. Athletics | F | 16 | $2505 \pm 431^{\text {a }}$ | $1.256 \pm .126$ | $11.1 \pm 2.5^{\text {a }}$ | $18.0 \pm 3.4^{\text {a }}$ | $3.9 \pm 0.8$ | $50.3 \pm 6.1^{\text {a }}$ | $17.7 \pm 1.4^{\text {a }}$ | $47.8 \pm 5.8^{\text {a }}$ | $22.5 \pm 3.2^{\text {a }}$ | $7.9 \pm 0.7^{\text {a }}$ |
|  | M | 11 | $3028 \pm 419$ | $1.328 \pm 0.117$ | $7.4 \pm 1.2$ | $10.4 \pm 1.4$ | $2.3 \pm 0.4$ | $63.9 \pm 4.6$ | $19.3 \pm 1.6$ | $60.9 \pm 4.3$ | $29.2 \pm 2.5$ | $8.8 \pm 0.9$ |
| 3. Basketball | F | 35 | $2620 \pm 502^{\text {a }}$ | $1.220 \pm 0.142^{\text {a }}$ | $17.1 \pm 4.4^{\text {a }}$ | $25.6 \pm 4.7^{\text {a }}$ | $5.5 \pm 1.3^{\text {a }}$ | $48.6 \pm 5.2^{\text {a }}$ | $15.8 \pm 1.4^{\text {a }}$ | $46.0 \pm 4.9^{\text {a }}$ | $20.2 \pm 2.6^{\text {a }}$ | $6.6 \pm 0.8^{\text {a }}$ |
|  | M | 45 | $3292 \pm 516$ | $1.299 \pm 0.140$ | $12.1 \pm 4.1$ | $14.8 \pm 3.8$ | $3.3 \pm 1.1$ | $68.5 \pm 8.1$ | $18.8 \pm 1.5$ | $65.2 \pm 7.7$ | $30.9 \pm 4.2$ | $8.5 \pm 0.8$ |
| 4. Fencing | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
| 5. Gymnastics | F | 12 | $2018 \pm 332$ | $1.117 \pm 0.107$ | $11.8 \pm 3.5$ | $22.7 \pm 4.9^{\text {a }}$ | $4.6 \pm 1.3$ | $38.7 \pm 4.3^{\text {a }}$ | $15.0 \pm 1.2^{\text {a }}$ | $36.6 \pm 4.1^{\text {a }}$ | $16.2 \pm 1.9^{\text {a }}$ | $6.3 \pm 0.5^{\text {a }}$ |
|  | M | $2^{\#}$ | $2472 \pm 740$ | $1.190 \pm 0.148$ | $7.8 \pm 4.0$ | $12.0 \pm 2.8$ | $2.8 \pm 1.3$ | $55.0 \pm 14.2$ | $19.7 \pm 4.5$ | $52.5 \pm 13.5$ | $24.5 \pm 7.6$ | $8.8 \pm 2.5$ |
| 6. Handball | F | $4{ }^{\text {\# }}$ | $2544 \pm 436^{\text {a }}$ | $1.270 \pm 0.107$ | $18.5 \pm 2.1^{\text {a }}$ | $27.3 \pm 1.6$ | $6.6 \pm 0.8^{\text {a }}$ | $49.2 \pm 3.1^{\text {a }}$ | $17.6 \pm 0.9^{\text {a }}$ | $46.6 \pm 2.8^{\text {a }}$ | $20.4 \pm 1.5^{\text {a }}$ | $7.3 \pm 0.5^{\text {a }}$ |
|  | M | 37 | $3342 \pm 486$ | $1.346 \pm 0.105$ | $13.6 \pm 5.9$ | $16.1 \pm 5.3$ | $4.0 \pm 1.7$ | $69.2 \pm 7.9$ | $20.5 \pm 1.6$ | $65.9 \pm 7.5$ | $31.1 \pm 4.0$ | $9.2 \pm 0.9$ |
| 7. Hockey Rink | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $2^{\#}$ | $2784 \pm 217$ | $1.222 \pm 0.066$ | $7.0 \pm 0.9$ | $9.8 \pm 1.7$ | $2.2 \pm 0.0$ | $64.2 \pm 4.6$ | $20.4 \pm 3.9$ | $61.5 \pm 4.8$ | $28.9 \pm 2.4$ | $9.2 \pm 1.9$ |
| 8. korfball | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
| 9. Modern Pentathlon | F | $2{ }^{\text {\# }}$ | $1915 \pm 118$ | $1.026 \pm 0.012$ | $10.2 \pm 1.5$ | $18.1 \pm 0.2^{\text {a }}$ | $3.7 \pm 0.2$ | $46.3 \pm 6.1$ | $16.6 \pm 0.6$ | $44.4 \pm 6.0$ | $19.3 \pm 3.0$ | $6.9 \pm 0.4$ |
|  | M | 5 | $2489 \pm 403$ | $1.159 \pm 0.133$ | $8.2 \pm 1.8$ | $12.3 \pm 2.2$ | $2.6 \pm 0.6$ | $57.6 \pm 5.6$ | $18.2 \pm 1.8$ | $55.1 \pm 5.2$ | $25.5 \pm 2.8$ | $8.0 \pm 0.9$ |
| 10. Motorsport | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
| 11. other combat sports | F | $4{ }^{\text {\# }}$ | $2184 \pm 518^{\text {a }}$ | $1.120 \pm 0.148$ | $15.9 \pm 1.8^{\text {a }}$ | $27.6 \pm 2.1^{\text {a }}$ | $5.8 \pm 0.7^{\text {a }}$ | $42.2 \pm 7.8^{\text {a }}$ | $15.2 \pm 1.9^{\text {a }}$ | $40.0 \pm 7.4^{\text {a }}$ | $17.9 \pm 4.6^{\text {a }}$ | $6.4 \pm 1.1^{\text {a }}$ |
|  | M | 13 | $2827 \pm 451$ | $1.261 \pm 0.118$ | $9.0 \pm 3.0$ | $12.9 \pm 3.2$ | $2.9 \pm 1.0$ | $59.9 \pm 7.4$ | $19.1 \pm 1.9$ | $57.1 \pm 7.0$ | $26.4 \pm 3.2$ | $8.4 \pm 0.8$ |
| 12. Rowing | F | $0^{\text {\# }}$ | 2209 | 1.072 | 19.8 | 28.5 | 6.8 | 49.6 | 17.1 | 47.3 | 21.2 | 7.3 |
|  | M | $6{ }^{\#}$ | $2861 \pm 207$ | $1.201 \pm 0.074$ | $10.8 \pm 1.9$ | $14.1 \pm 2.2$ | $3.2 \pm 0.6$ | $65.4 \pm 3.2$ | $19.1 \pm 1.0$ | $62.5 \pm 3.1$ | $29.3 \pm 1.5$ | $8.6 \pm 0.4$ |
| 13. Rugby | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | 39 | $3364 \pm 425$ | $1.381 \pm 0.110$ | $17.1 \pm 9.3$ | $18.5 \pm 6.8$ | $5.2 \pm 2.9$ | $70.1 \pm 10.0$ | $21.3 \pm 2.8$ | $66.7 \pm 9.6$ | $31.3 \pm 4.5$ | $9.5 \pm 1.3$ |
| 14. Sailing | F | $0^{\#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | $4^{\#}$ | $2902 \pm 241$ | $1.246 \pm 0.038$ | $9.1 \pm 2.1$ | $11.8 \pm 1.9$ | $2.7 \pm 0.6$ | $66.7 \pm 4.3$ | $20.3 \pm 1.1$ | $63.8 \pm 4.0$ | $29.5 \pm 1.8$ | $8.9 \pm 0.4$ |

Table 7.6. (cont.) Body composition for the Dual-energy X-ray absorptiometry outputs.

| Sport | gender | n | WB BMC (g) | WB BMD $\left(\mathrm{g} / \mathrm{cm}^{2}\right)^{b, c}$ | WB FM (kg) ${ }^{\mathrm{b}, \mathrm{c}}$ | WB FM (\%) b, c | $\underset{\mathrm{b}, \mathrm{c}}{\mathrm{FMI}}\left(\mathbf{k g} / \mathrm{m}^{2}\right)$ | WB FFM $(\mathbf{k g})^{b, c}$ | $\begin{aligned} & \text { FFMI } \\ & \left(\mathbf{k g} / \mathrm{m}^{2}\right)^{\mathrm{b}, \mathrm{c}} \end{aligned}$ | WB LST (kg) <br> b, c | ALST (kg) ${ }^{\text {b, }}$ c | $\begin{aligned} & \text { ALSTI } \\ & \left(\mathbf{k g} / \mathbf{m}^{2}\right)^{b, c} \end{aligned}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 15. Soccer | F | $0{ }^{\# \#}$ |  |  |  |  |  |  |  |  |  |  |
|  | M | 28 | $2988 \pm 328$ | $1.341 \pm 0.090$ | $8.8 \pm 2.5$ | $12.1 \pm 2.7$ | $2.8 \pm 0.7$ | $63.2 \pm 5.6$ | $20.4 \pm 1.5$ | $60.2 \pm 5.3$ | $28.1 \pm 2.6$ | $9.1 \pm 0.8$ |
| 15. Surf | F | 1 | 1664 | 1.045 | 10.1 | 21.8 | 4.5 | 36.4 | 16.2 | 34.7 | 15.0 | 6.7 |
|  | M | 0 |  |  |  |  |  |  |  |  |  |  |
| 16. Swimming | F | 22 | $2112 \pm 268^{\text {a }}$ | $1.095 \pm 0.067^{\text {a }}$ | $13.9 \pm 2.4^{\text {a }}$ | $23.3 \pm 2.6^{\text {a }}$ | $5.0 \pm 0.8^{\text {a }}$ | $45.4 \pm 5.1^{\text {a }}$ | $16.3 \pm 1.0^{\text {a }}$ | $43.3 \pm 4.9^{\text {a }}$ | $18.8 \pm 2.2^{\text {a }}$ | $6.8 \pm 0.4{ }^{\text {a }}$ |
|  | M | 36 | $2599 \pm 460$ | $1.167 \pm 0.126$ | $8.9 \pm 3.2$ | $12.5 \pm 3.4$ | $2.8 \pm 0.9$ | $61.0 \pm 7.3$ | $19.0 \pm 1.5$ | $58.4 \pm 7.0$ | $26.7 \pm 3.3$ | $8.3 \pm 0.7$ |
| 17. Tennis | F | $5{ }^{\text {\# }}$ | $2180 \pm 330^{\text {a }}$ | $1.136 \pm 0.139^{\text {a }}$ | $16.4 \pm 1.6^{\text {a }}$ | $26.2 \pm 2.0^{\text {a }}$ | $5.8 \pm 0.6{ }^{\text {a }}$ | $46.6 \pm 6.7^{\text {a }}$ | $16.5 \pm 1.6^{\text {a }}$ | $44.5 \pm 6.4^{\text {a }}$ | $19.3 \pm 2.6^{\text {a }}$ | $6.8 \pm 0.6^{\text {a }}$ |
|  | M | 11 | $2572 \pm 322$ | $1.197 \pm 0.096$ | $12.7 \pm 6.1$ | $17.1 \pm 5.6$ | $4.1 \pm 1.9$ | $59.6 \pm 8.3$ | $19.0 \pm 1.9$ | $57.0 \pm 8.1$ | $26.8 \pm 4.3$ | $8.5 \pm 1.0$ |
| 18. Triathlon | F | 10 | $1947 \pm 189^{\text {a }}$ | $1.068 \pm 0.065^{\text {a }}$ | $11.4 \pm 4.5^{\text {a }}$ | $20.0 \pm 6.1^{\text {a }}$ | $4.2 \pm 1.9^{\text {a }}$ | $44.7 \pm 2.7^{\text {a }}$ | $16.1 \pm 1.3^{\text {a }}$ | $42.7 \pm 2.6^{\text {a }}$ | $18.8 \pm 1.6^{\text {a }}$ | $6.8 \pm 0.7^{\text {a }}$ |
|  | M | 38 | $2448 \pm 292$ | $1.153 \pm 0.091$ | $7.7 \pm 1.4$ | $11.9 \pm 2.0$ | $2.5 \pm 0.5$ | $57.4 \pm 4.2$ | $18.5 \pm 1.1$ | $55.0 \pm 4.0$ | $25.1 \pm 2.2$ | $8.1 \pm 0.6$ |
| 19. Volleyball | F | 16 | $2518 \pm 390^{\text {a }}$ | $1.207 \pm 0.097^{\text {a }}$ | $17.4 \pm 5.6^{\text {a }}$ | $25.6 \pm 4.7^{\text {a }}$ | $5.6 \pm 1.4^{\text {a }}$ | $49.6 \pm 6.9^{\text {a }}$ | $16.2 \pm 1.4^{\text {a }}$ | $47.1 \pm 6.6^{\text {a }}$ | $21.1 \pm 3.6^{\text {a }}$ | $6.9 \pm 0.8^{\text {a }}$ |
|  | M | 17 | $3793 \pm 478$ | $1.399 \pm 0.101$ | $12.9 \pm 3.3$ | $14.3 \pm 3.4$ | $3.4 \pm 0.9$ | $76.7 \pm 6.9$ | $20.2 \pm 1.4$ | $72.9 \pm 6.6$ | $35.0 \pm 3.9$ | $9.2 \pm 0.8$ |
| 20. Wrestling + Judo | F | 15 | $2390 \pm 402^{\text {a }}$ | $1.238 \pm 0.089^{\text {a }}$ | $13.6 \pm 4.5^{\text {a }}$ | $23.0 \pm 6.0^{\text {a }}$ | $5.1 \pm 1.6^{\text {a }}$ | $44.2 \pm 6.9^{\text {a }}$ | $16.7 \pm 2.0^{\text {a }}$ | $41.8 \pm 6.6^{\text {a }}$ | $18.1 \pm 3.3^{\text {a }}$ | $6.8 \pm 1.0^{\text {a }}$ |
|  | M | 45 | $3038 \pm 412$ | $1.365 \pm 0.128$ | $8.7 \pm 3.0$ | $12.2 \pm 3.0$ | $2.9 \pm 1.0$ | $61.2 \pm 7.8$ | $20.7 \pm 2.3$ | $58.2 \pm 7.5$ | $26.6 \pm 3.6$ | $9.0 \pm 1.1$ |
| All sample | F | 143 | $2347 \pm 457^{\text {a }}$ | $1.177 \pm 0.127^{\text {a }}$ | $14.6 \pm 4.5^{\text {a }}$ | $23.5 \pm 5.1^{\text {a }}$ | $5.1 \pm 1.4^{\text {a }}$ | $22.6 \pm 3.1^{\text {a }}$ | $16.3 \pm 1.5^{\text {a }}$ | $44.2 \pm 6.0^{\text {a }}$ | $19.6 \pm 3.1^{\text {a }}$ | $6.8 \pm 0.8^{\text {a }}$ |
|  | M | 339 | $3022 \pm 556$ | $1.291 \pm 0.141$ | $10.9 \pm 5.5$ | $13.9 \pm 4.6$ | $3.3 \pm 1.6$ | $30.4 \pm 4.2$ | $19.8 \pm 2.1$ | $61.6 \pm 8.5$ | $28.7 \pm 4.4$ | $8.8 \pm 1.0$ |
| Sport Comparisons | F |  | $\begin{aligned} & { }^{\text {b) }} 18 \neq 2,19,3 ; \\ & 5,16 \neq 3 \end{aligned}$ | $\begin{aligned} & { }^{\text {b) }} 18 \neq 20,2 ; 16 \\ & \neq 3,20,2 \end{aligned}$ | $\begin{aligned} & \text { b) } 2 \neq 3,19, \\ & 6 ; 18,5 \neq 3 \end{aligned}$ | $\begin{aligned} & { }^{\text {b) }} 2 \neq 3,19,17, \\ & 6,11 \end{aligned}$ | ${ }^{\text {b) }} 2 \neq 3,19,6$ | ${ }^{\text {b) }} 5 \neq 3,19,2$ | ${ }^{\text {b) }} 5,3 \neq 2$ | ${ }^{\text {b) }} 5 \neq 3,19,2$ | $\begin{aligned} & { }^{\text {b) }} 5 \neq 3,19,2 ; \\ & 20,16 \neq 2 \end{aligned}$ | $\begin{aligned} & { }^{\text {b) }} 5,3,16,19 \neq \\ & 2 \end{aligned}$ |
|  | M |  | $\begin{aligned} & { }^{\text {c) }} 18 \neq 15 ; 18, \\ & 16 \neq 20,3,6, \\ & 13,19 ; 9,11 \neq \\ & 19 ; 17 \neq 3,6, \\ & 13,19 \end{aligned}$ | $\begin{aligned} & \text { c) } 18 \neq 2 ; 18,16 \\ & \neq 3,15,6,20, \\ & 13,19 ; 17 \neq 20, \\ & 13,19 \end{aligned}$ | $\begin{aligned} & \text { c) } 2,18,20, \\ & 15,16 \neq 3 \\ & 19,6,13 ; 18 \\ & \neq 17 ; 11 \neq 13 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 2 \neq 3,6,17, \\ & 13 ; 18 \neq 6,13 ; \\ & 15,16 \neq 13 ; \\ & 20 \neq 3,6,13 \end{aligned}$ | $\begin{aligned} & \text { c) } 2 \neq 3,6,17, \\ & 13 ; 18 \neq 3,6, \\ & 13 ; 16 \neq 6,13 ; \\ & 15 \neq 13 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 18,20,16 \neq \\ & 3,6,13,19 ; \\ & 17,9,11,15 \neq \\ & 19 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 18,3 \neq 15,6, \\ & 20,13 ; 16 \neq 6, \\ & 20,13 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 18,20,16 \neq \\ & 3,6,13,19 ; 9, \\ & 17,11,15 \neq \\ & 19 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 18,20,16 \neq \\ & 3,6,13,19 ; 9, \\ & 17,15 \neq 19 ; \\ & 11 \neq 3,6,19 \end{aligned}$ | $\begin{aligned} & { }^{\text {c) }} 18 \neq 20,15 \\ & 19,6,13 ; 16 \neq \\ & 15,6,13 ; 3 \neq \\ & 6,13 \end{aligned}$ |

[^6]${ }^{\text {a) }}$ Significant different from males ( $p<0.05$ ); ${ }^{\text {b) }}$ Significant differences between sports for females ( $p<0.05$ ); ${ }^{\text {c) }}$ Significant differences between sports for males ( $p<0.05$ )

Significant differences were observed between sexes for almost all variables and the differences varied by sport. In addition some differences in the means across sports were found for all variables. Regarding anthropometry parameters (Table 4), female gymnasts presented lower weight and height whereas basketball and volleyball had higher values. In the $\sum 7$ SKF female athletics had lower values compared to soccer, volleyball, judo and wrestling, basketball, tennis and archery and shooting. In males triathlon and gymnastics were heavier than hockey rink, rowing, basketball, handball, volleyball, and rugby athletes. Generally volleyball, basketball, handball, rowing, athletics, swimming, and sailing male athletes were taller than gymnasts and practitioners of judo. Concerning the $\sum 7$ SKF, differences were observed between athletics and triathlon compared to basketball, hockey rink, handball, sailing, archery and shooting, and rugby that presented higher values in this parameter.

In DXA variables (Table 5) we also found differences across sports in the distributions. Regarding BMD we found in both male and female triathlon and swimming athletes, lower BMD values whereas athletics, judo and wrestling females and basketball, soccer, handball, judo and wrestling, rugby, and volleyball male athletes had the highest values. In athletics the lower \%FM was found in contrast to basketball, volleyball, tennis, and handball male and female athlete. Also in female of other combat sports higher $\% \mathrm{FM}$ results were found. We further verified that in females, basketball, volleyball, and athletics present higher values in ALST compared to gymnastics. In male team sports such as basketball, handball, rugby, and volleyball, higher ALST was observed compared to triathlon, judo and swimming.

It is important to highlight that for both sex and sport comparisons the p-values are unadjusted for multiple comparisons, keeping in mind the large number of outcomes and sports considered for each sex.

We further analyzed how the anthropometric variables were associated with DXA parameters, and verified strong associations between the $\Sigma 7$ SKF and $\% \mathrm{FM}$ (females: $\mathrm{r}=0.80, \mathrm{p}<0.001$; males: $\mathrm{r}=0.88 ; \mathrm{p}<0.001$ ). The ALST was explained by all the muscle circumferences in the same model (females: $\mathrm{r}=0.77$, $\mathrm{p}<0.001$; males: $\mathrm{r}=0.79$; $\mathrm{p}<0.001$ ).

### 7.5. Discussion

Despite the recognized importance of body composition for athletic performance [1-3] and health [9], appropriate reference values for the athletic population have been lacking. In this study we developed sex and sport specific percentiles for body composition at a molecular and whole-body level, using dual-energy X-ray absorptiometry (DXA) and anthropometry, respectively.

DXA is a widespread method for the assessment of the athletic population has it permits the acquisition of regional in addition to total body composition, avoiding the use of costly and scarce medical imaging techniques [1, 12]. Concerning DXA outcomes we presented the $5^{\text {th }}, 25^{\text {th }}, 50^{\text {th }}, 75^{\text {th }}$, and $90^{\text {th }}$ percentile for total (whole-body scan) and regional (sub-total, trunk, and appendicular) body composition, including measures of BMD, BMC, absolute and percent FM, FFM, and LST. Additionally we also presented reference percentiles for height-normalized indexes as BMI by dividing FM, FFM, and ALST by height squared. This approach has been suggested to allow comparisons between individuals [24], in addition reference values for the adult population have already been developed [16]. The percentiles for DXA measurements are presented in Tables S17 to S40. In order to correctly interpret these percentiles we give an example for a male handball player that has a fat mass (FM) of $13.0 \%$. By Table S21 we can see that the estimate for the $5^{\text {th }}$ percentile is $8.0 \%$ ( $95 \% \mathrm{CI}: 3.3-$ $11.7 \%$ ), the estimate for the $25^{\text {th }}$ percentile is $12.9 \%(95 \% \mathrm{CI}: 9.7-15.7 \%)$ and the estimate for the $50^{\text {th }}$ percentile is $16.3 \%(95 \% \mathrm{CI}: 14.2-18.5 \%)$. Thus we can say with $95 \%$ certainty that the male handball player falls in the bottom half of the distribution but is not in the lowest $5 \%$ of that distribution; we estimate that his FM measurement is at about the $25^{\text {th }}$ percentile for his sport.

Athletes as a rule have a lower \% FM than nonathletes of the same chronologic age [3]. The excess of FM may have a negative impact on sports performance and is often viewed as a major limiting factor in athletic achievements [3]. On the other hand in the athletic setting a lower percent FM may be related to several heath complications that sports professionals must be aware [9]. Malina et al. [3] has made a review for estimated fatness of athletes from different sports, however the majority of the results were based on densitometric methods to estimate FM (e.g. hydrostatic weighting, air
displacement pletysmography or anthropometry prediction equations). The variability in fat-free mass density is one of the main factors limiting the accuracy of body composition estimates using densitometric models [25] and in the athletic population it has been observed that deviations from the established value of $1.1 \mathrm{~g} / \mathrm{cc}$ may occur [26]. In this investigation we developed reference percentiles using DXA, a 3-compartment method that provides a reliable and valid alternative over other recognized methods due to its speed and convenience [12].

It has been investigated that athletes present a much larger FFM than nonathletes. Although not entirely, the majority of these differences may be related to an increased skeletal mass [19]. A relatively large fraction of total body skeletal mass is in the appendages and a high percentage of ALST is skeletal mass, thus estimation of ALST by DXA is a potentially practical and accurate method of quantifying human skeletal mass in vivo [27, 28]. On the other hand, ALST divided by height ${ }^{2}$ (ALSTI) has been suggested as a proxy index for sarcopenia [29]. It is expected that an athletic population, particularly those engaged in high intensity training present an increased skeletal mass compared to non-athletes [30]. In this study, in addition to whole-body FFM and LST we presented quantiles for both ALST and ALSTI, for each sex and separately for several sports that may present differences in their body composition profile.

Another advantage of using DXA in the athletic population is its ability to assess BMD. The position stand of the American College of Sports Medicine (ACSM) regarding the female athlete triad syndrome (FTS) refers that BMD should be assessed with DXA whenever the athlete presents a history of stress fractures or fractures from minimal trauma [9]. In our study we observed that the lower BMD was observed among triathlon and swimming athletes. FTS is more often seen in endurance sports like triathlon along with sports that emphasize thinness (e.g. gymnastics, figure skating and dancing). On the other hand it has been suggested that a higher peak bone mass may be achieved by regularly performing weight bearing exercises, particularly if associated with impacts [31]. In fact we observed that swimmers, a nonweight bearing sport, presented a lower BMD compared to other sports with more impact.

Despite all the advantages, DXA may not be practical for field assessment and caution is necessary when using this method on several occasions, perhaps no more than four times per year [1], not only due to the cumulative radiation dose but also because of the error of measurement in detecting small body composition changes [32] may lead to misinterpretation of data. Anthropometry provides a simple, relative inexpensive and non-invasive field method for estimating body composition [1, 33]. At this regard we also presented percentiles for anthropometry outcomes (Tables S1 to S16). The interpretation of the percentiles for these variables is similar to the example given for \%FM from DXA.

When using anthropometry, inconsistency exists when using anthropometry to estimate molecular body composition compartments, particularly in the athletic population as the equations rely on assumptions that may not be valid in athletes [26, 34]. The other source of error is the lack of standardization for the measurement of skinfolds and circumferences [1]. In order to solve these common issues we presented anthropometric data by using a standardized protocol [19]. On the other hand using the raw data instead of applying an equation, i.e the use of sum of skinfolds ( $\sum 7 \mathrm{SKF}$, $\sum_{\text {app }}$ SKF, $\sum_{\text {trunk }}$ SKF, $\sum_{\text {arm }}$ SKF, and $\sum_{\text {leg }}$ SKF) and body circumferences (Hip, arm, midthigh, calf, and abdominal circumferences), we were able to reduce errors by avoiding assumptions that may not be adequate in the athletic population. At this regard Ackland et al. (2012) [1] and Marfell-Jones (2001) [35] highlighted the importance of individual and sum of skinfolds thickness in its own as a valid proxy measurement of adiposity. In our investigation we observed that the $\Sigma 7$ SKF were highly associated with FM from DXA in both sexes.

Despite the fact that circumferences are not often used to assess body composition in the athletic population, it seems likely that muscle circumferences (Mc) may be a useful anthropometric tool as a representation of skeletal muscle [36]. In this study, following the procedure suggested by Heymsfield et al. [21], we included reference quantiles for arm, thigh, and calf Mc in addition to unadjusted measures for the skinfold thickness. Besides we were able to verify that muscle circumferences were associated with ALST, which is a predictor of whole-body skeletal muscle [27]. It is important to highlight that anthropometry requires adequate training by an experienced
professional and quality control, including analyses of reliability data and calibration of equipments [33].

### 7.5.1 Study limitations:

The DXA reference values presented in this study are only comparable with those from Hologic fan beam DXA scanner (software version 12.4 or higher).

Due to the small sample size in each sport we were not able to present position specific reference percentiles for each body composition outcome. Moreover, the athletics sport was only comprised by sprinters, hurdlers, and jumpers (long and triple jump).

Ethnic variation should also be considered as a limitation of this study given that variation exists in body proportions and composition [37].

### 7.6. Conclusions

This study provides reference body composition percentiles for sex and sport athletes. Sports professionals will benefit from using this tool for assessing and classifying body composition in athletes. We presented total and regional body composition reference by using Dual-energy X-ray absorptiometry. In addition, given its applicability in the field setting we also developed reference percentiles for wholebody composition by using anthropometric methods (sum of skinfolds, circumferences, and muscle circumferences). These reference values should be helpful in the evaluation of athletes from different sports not only regarding performance but also health-related criteria, and to establish directions for research purposes.

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### 8.1. Overview

Assessing body composition in the athletic field plays an important role in monitoring athletic performance and training regimens as well as the health status of the athlete [1]. In the literature review section a comprehensive model, proposed by Wang et al. [2] for organizing human body composition was examined. In this systematization, body mass can be viewed as five distinct and separate but integrated levels of increasing complexity. The five levels are I, atomic; II, molecular; III, cellular; IV, tissue-system; and V , whole body. In line with this, body-composition research includes three interconnecting areas: body composition rules, body composition methodology, and body composition alterations. In the body composition rules the proportions of various components and their steady-state associations among the atomic, molecular, cellular, tissue system, and whole-body levels were explored. In the second area, the methodology, we analysed the main in vivo methods that can be used to estimate body composition in each of the five levels of body composition [2]. In the body composition alteration the influences of biological factors on various levels and components, such as growth, aging, and physical activity were examined, with a particular focus on physical activity related energy expenditure, since this is of major interest in the athletic field. Finally, we included a section where we analysed each of the three body composition areas in athletic populations. The presentation of our main research findings and the practical applications of our investigation will follow the same rational (figure 8.1).

An exhaustive discussion of each of the four studies' main findings was included in the respective chapters. The rationale of this section was to gather and integrate the contributions of the four studies, by summarizing the main results and globally reflecting on the implications for future research and practical applications. Limitations of these studies and future research avenues are also disclosed.


Figure 8.1. Interconnection between the three body composition research areas and the studies from the present dissertation

### 8.2. Main research findings

### 8.2.1. BODY COMPOSITION RULES

In the present dissertation we explored the applicability of the rules that are currently used for the molecular level of analysis in athletic populations (Chapter 6: study 3). At the molecular level constant densities and composition of the FFM components are assumed. Fat-mass is assumed to present a density of $0.9007 \mathrm{~g} / \mathrm{cm}^{3}$, whereas for FFM is assumed to be $1.1000 \mathrm{~g} / \mathrm{cm}^{3}$, given the constant composition and densities that are assumed for the FFM components (water, mineral, and protein). We verified that in elite junior male and female basketball players the composition of the FFM differed from the established values (TBW/FFM $=73.8 \%$; M/FFM $=6.8 \%$; $\operatorname{Prot} / \mathrm{FFM}=19.4 \%$ ) both at a pre-season, when athletes came from an 8 -week resting period, without any basketball practice and at a competitive period, close to the main National competition, where athletes are deemed to be at their best playing performance. In both male and females the water and mineral fractions of FFM were lower than the assumed values while the protein fraction was considerably higher.

Regardless, in males the $\mathrm{FFM}_{\mathrm{D}}$ was not different than $1.1000 \mathrm{~g} / \mathrm{cm}^{3}$. On the other hand, the females presented a higher $\mathrm{FFM}_{\mathrm{D}}$ at the competitive period assessment, probably due to a decreased in the water FFM fraction. Other investigations [3-6] verified that the assumptions that are made regarding the FFM composition and density may not be valid in athletes. However, to our knowledge this was the first investigation that verified these assumptions in team sports athletes using a longitudinal approach. Silva et al. [6] also assessed athletes at two different periods of the season, particularly male judo athletes from a period of weight stability to prior a competition. Some of these athletes intentionally lost weight between assessments and therefore variations in body composition were expected, particularly regarding the FFM hydration. The basketball players assessed in study 3 (chapter 6) were not intentionally modifying their body composition therefore our findings suggest that in athletes, violations of the composition and density of FFM may exist in team sport players, which are not categorized as weight-sensitive sports [1].

### 8.2.2. BODY COMPOSITION METHODOLOGY

The primary focus of the scientific community has been the assessment of the molecular level of body composition analysis, particularly the assessment of FM and FFM [7]. Dual-energy X-ray absorptiometry measurement has been suggested as a valid alternative to reference methods in athletic populations due to its speed (fan-beam densitometers), but also because the measurement is minimally influenced by water fluctuation [1, 8, 9]. Regardless one of its major limitations relates to the scan area. DXA systems are not capable of assessing individuals taller than the scan area, which varies between 185 and 197 cm , depending on the equipment [10]. This limitation affects particularly athletes involved in sports where height is a major factor of performance, such as basketball and volleyball. In the first study (chapter 4) we proposed an alternative to solve this methodological limitation in a fan-beam densitometer, particularly by suggesting the sum of a head and a trunk plus limbs scan. Other researchers had suggested approaches to solve this limitation [11, 12] but for pencil-beam densitometers. The new approach that was suggested in study 1 (chapter 4) allowed the assessment of athletes for study 2 (chapter 5), study 3 (chapter 6), and study 4 (chapter 7) using dual-energy X-ray absorptiometry to characterize body composition
(studies 2 and 4) and to estimate body composition components that allowed the application of a 4-component model at the molecular level, the estimation of body cell mass at the cellular level, and the prediction of the skeletal muscle mass at the tissuesystem level (study 3). Despite the convenience of using DXA in athletic populations, to date no reference values for the assessed body components existed for athletic populations. Kelly et al. [13] have presented reference values for body composition determined by DXA but for the general US population. In study 4 (chapter 7) we were able to provide percentiles for whole-body and regional body composition for athletes from different sports and according to sex. These reference percentiles should be a helpful tool for sports professionals to prescribe an adequate exercise training and dietary intake over the season.

In the study 3 (chapter 6), as already stated above, we questioned the assumptions that are made at the molecular level regarding the density and composition at the FFM, which are the cornerstones of 2-componet molecular methods [14]. Therefore, we reinforced that at this level densitometric and hydrometric methods must be used with caution to assess body composition in athletes. In fact, at the field setting the majority of the investigation have been conducted using either anthropometric measures to estimate molecular components [15-17] or bio-impedance analysis [18] developed against densitometric (for anthropometric based equations) or hydrometric techniques (BIA based equations) that lied on the aforementioned assumptions regarding the FFM density and hydration. Despite these consequences, anthropometric methods have a widespread utility for monitoring athletes by providing a simple and highly portable method for estimating body composition [1]. At this regard the use of raw anthropometric variables has been suggested (e.g. sum of skinfolds) [19, 20]. In fact, in study 3 (chapter 6) we verified that raw anthropometric measures were associated with molecular [the sum of seven skinfolds ( $\Sigma 7 \mathrm{SKF}$ ) was associated with FM] and tissue [the muscle circumferences (Mc) were associated with the skeletal muscle mass] components. In the same study (study 3, chapter 6) we verified that the $\sum 7$ SKF was associated not only with performance measures [handgrip and countermovement jump (CMJ)] in cross sectional assessments but also its reduction over a season was related to an improved performance. Similar conclusions were observed for the use of arm and thigh muscle circumferences given that they explained
the handgrip and the CMJ, whereas changes in the calf muscle circumference were correlated with the CMJ test. Since raw anthropometric variables were associated with components from other body composition levels (i.e. molecular and tissue level) and with athletic performance (study 3, chapter 6), and given that anthropometric variables provide a simple and practical tool for body composition assessment in the field setting we provided reference values for athletes at the whole body level (study 4, chapter 7). In fact, Marfel-Jones et al. [20] has suggested that investigators should collate the large amounts of skinfold data to publish skinfold sum norms. Therefore, in study 4 (chapter 7) we provided percentiles for sum of skinfolds (whole body, arms, legs, trunk, and appendicular sums of skinfolds) and also for circumferences and muscle circumferences. These percentiles provide a reference for data comparison that will allow a better understanding of the raw data anthropometric variables among sport professionals and athletes.

### 8.2.3. BODY COMPOSITION ALTERATIONS

Several factors are recognized to influence body composition alterations. The regulation between energy intake and energy expenditure, is a major determinant of changes in body components. Athletes need to consume adequate energy to maintain a healthy body composition profile but also to maximize training effects [21]. Many athletes are chronically energy deficient and it is of extreme importance to characterize athletes' energy expenditure in order to identify individual energy requirements [22]. The study 2 (chapter 5) aimed to validate an objective measure of physical activity that combines an heart rate monitor with a motion sensor in a sample of male and female basketball players. However we observed that the device did not provide accurate measurements of individual energy expenditure, which limits the use of this device in athletes. Other research studies have already shown the inaccuracies associated with energy expenditure assessment by electronic devices that combine objective measures of physical activity in athletic populations [23]. At this point future research needs to focus not only in validating existing physical activity monitors but also developing new algorithms specific for the athletic population.

In study 4 (chapter 7) we established molecular and whole-body composition reference values for athletes of different sports Therefore athletes may now have
standard values for defining an appropriate goal for their body composition profile. In order to achieve that goal it is determinant to accurately estimate energy expenditure to adequately estimate their energy requirements, which imposes the development and validity of new physical activity technologies

Regarding body composition alterations, seasonal variations are expected in the course of a sports season [16, 24-27]. The majority of the investigations regarding body composition alterations during a season have focused on the whole body and the molecular levels, while most of the investigators focus on FM using 2-component models. Therefore, in study 3 (chapter 6) we analyzed body composition changes from the pre-season to the main competitive period in elite junior basketball players by scrutinising 4 different levels of body composition (i.e. molecular, cellular, tissuesystem, and whole body). In this study we verified that in both sexes enhancements in body composition occur in each of the four levels that were analyzed. Alterations in body composition have been investigated to impact performance [16, 25, 26, 28]. In study 3 (chapter 6) we were also able to verify that body composition changes in different levels (i.e. molecular, cellular, tissue-system, and whole body) were related to changes in athletic performance in elite junior basketball players. In this study (study 3, chapter 6) we concluded that, despite the fact that components assessed in each of the four levels were cross-sectionally associated with performance tests (handgrip and CMJ), only whole-body and molecular variables explained changes in performance from the pre-season to the main competitive period.

### 8.3. Practical implications and future directions

In this section we summarized the practical findings derived from the studies to the real-world of sports and exercise settings.

### 8.3.1 BODY COMPOSITION RULES

- This investigation reinforced that the assumed density and composition of FFM at the molecular level may not be valid in athletes which limits the use of densitometric and hydrometric methods in the athletic population (study 3, chapter 6).


### 8.3.2. BODY COMPOSITION METHODOLOGY

- Although dual-energy X-ray absorptiometry provides an accurate measurement of body composition, it presented limitations when evaluating taller participants. In order to solve this methodological limitation, in study 1 (chapter 4) we proposed the sum of two separated scans: head scan and trunk plus limbs scan, to assess bone mineral content, fat mass, and lean mass, using a whole-body scan to accurately determine body composition in taller athletes and non-athletes. Our new proposed technique allows DXA to be used in individuals taller than the scan area for future investigations.
- In study 3 (chapter 6) we observed that the body composition obtained at the whole-body level, particularly by using raw anthropometric variables, may help coaches to easily monitor their athletes in the field. Indeed, we observed that simple field measures such as the sum of skinfolds or muscle circumferences are associated with improvements in basketball players' performance.


### 8.3.3 BODY COMPOSITION ALTERATIONS

- Energy expenditure assessment in the athletic field is of extreme importance for accurately estimate athletes' energy requirements. In study 2 (chapter 5) we verified that a combined heart rate and motion sensor is not valid to determine individual energy requirements in athletes. It is therefore emerging the need for developing new algorithms for existing devices specific for the athletic population.
- In study 3 (chapter 6) we presented data for basketball players that are sportspecific and might be a useful standard tool for comparison of body composition data in the course of a season in male and female junior basketball players. We observed that the season was associated with significant changes at the molecular, cellular, tissue, and whole-body level of body composition along with an improved handgrip strength and vertical jump. We also highlighted the relevance of tracking body composition given its association with performance in specific tests.
- In study 4 (chapter 7) we provided reference body composition percentiles ( $5^{\text {th }}$, $25^{\text {th }}, 50^{\text {th }}, 75^{\text {th }}$, and $95^{\text {th }}$ ) at the molecular and whole body composition levels. Sports professionals will benefit from our data by having the opportunity of defining a sex and sports specific goal for their body composition profile. However future research is still
required to increment the number of sports, specifically for the DXA-based molecular components and parameters for the available percentiles.


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## APPENDICES

Supplementary tables from chapter 7

Table S1 - Body weight (kg) percentiles by sport and sex

| Sport | Low | 0.05 <br> Estimate | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low |  | High | Low | 0.95 <br> Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 42.49 | 48.54 | 53.56 | 50.43 | 54.80 | 58.76 | 55.94 | 59.16 | 62.37 | 59.56 | 63.51 | 67.89 | 64.75 | 69.77 | 75.82 |
| Basketball | 47.24 | 54.32 | 60.25 | 57.73 | 62.81 | 67.42 | 65.03 | 68.71 | 72.40 | 70.00 | 74.61 | 79.69 | 77.17 | 83.11 | 90.18 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 35.66 | 43.30 | 49.45 | 43.87 | 49.42 | 54.36 | 49.58 | 53.67 | 57.77 | 52.99 | 57.93 | 63.48 | 57.90 | 64.05 | 71.69 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | 37.76 | 48.23 | 56.53 | 46.73 | 54.51 | 61.40 | 52.97 | 58.87 | 64.77 | 56.35 | 63.24 | 71.01 | 61.21 | 69.52 | 79.98 |
| Modern Pentathlon | 35.92 | 48.29 | 58.09 | 46.60 | 55.76 | 63.86 | 54.03 | 60.95 | 67.87 | 58.04 | 66.14 | 75.30 | 63.81 | 73.61 | 85.98 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 39.87 | 48.44 | 55.32 | 48.50 | 54.77 | 60.35 | 54.50 | 59.17 | 63.84 | 57.99 | 63.57 | 69.84 | 63.02 | 69.90 | 78.47 |
| Rowing | 43.62 | 54.80 | 63.68 | 52.81 | 61.17 | 68.59 | 59.20 | 65.60 | 72.01 | 62.61 | 70.03 | 78.39 | 67.53 | 76.41 | 87.58 |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 41.64 | 48.98 | 54.95 | 50.05 | 55.37 | 60.14 | 55.89 | 59.82 | 63.74 | 59.50 | 64.26 | 69.59 | 64.69 | 70.66 | 78.00 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 43.45 | 49.61 | 54.68 | 50.91 | 55.38 | 59.41 | 56.09 | 59.39 | 62.69 | 59.37 | 63.40 | 67.88 | 64.10 | 69.17 | 75.33 |
| Tennis | 43.34 | 53.20 | 61.08 | 52.27 | 59.58 | 66.06 | 58.49 | 64.01 | 69.53 | 61.95 | 68.44 | 75.74 | 66.94 | 74.82 | 84.68 |
| Triathlon | 37.38 | 47.31 | 55.20 | 46.46 | 53.77 | 60.25 | 52.77 | 58.26 | 63.76 | 56.28 | 62.75 | 70.07 | 61.33 | 69.21 | 79.14 |
| Volleyball | 40.29 | 52.05 | 61.45 | 52.57 | 61.08 | 68.63 | 61.10 | 67.36 | 73.62 | 66.09 | 73.64 | 82.15 | 73.28 | 82.67 | 94.43 |
| Wrestling and Judo | 37.48 | 46.11 | 53.10 | 47.80 | 54.01 | 59.54 | 54.97 | 59.51 | 64.01 | 59.45 | 65.01 | 71.18 | 65.88 | 72.91 | 81.51 |
|  |  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |
| Archery and Shooting | 41.70 | 58.63 | 71.01 | 55.50 | 67.45 | 77.55 | 65.08 | 73.59 | 82.09 | 69.63 | 79.72 | 91.68 | 76.17 | 88.55 | 105.48 |
| Athletics | 55.77 | 62.83 | 68.53 | 64.40 | 69.44 | 73.92 | 70.41 | 74.03 | 77.66 | 74.15 | 78.62 | 83.66 | 79.53 | 85.23 | 92.29 |
| Basketball | 57.13 | 65.17 | 71.87 | 69.26 | 74.95 | 80.09 | 77.70 | 81.76 | 85.81 | 83.42 | 88.56 | 94.25 | 91.65 | 98.34 | 106.38 |
| Fencing | 47.92 | 60.40 | 69.73 | 58.98 | 67.81 | 75.35 | 66.67 | 72.96 | 79.26 | 70.57 | 78.11 | 86.94 | 76.20 | 85.52 | 98.00 |
| Gymnastics | 44.43 | 53.99 | 61.43 | 54.58 | 61.33 | 67.22 | 61.63 | 66.44 | 71.24 | 65.66 | 71.54 | 78.29 | 71.45 | 78.88 | 88.44 |
| Handball | 56.62 | 66.37 | 74.29 | 69.69 | 76.56 | 82.69 | 78.77 | 83.65 | 88.53 | 84.61 | 90.74 | 97.61 | 93.01 | 100.93 | 110.68 |
| Hockey Rink | 55.33 | 61.73 | 67.07 | 64.97 | 69.52 | 73.64 | 71.67 | 74.94 | 78.21 | 76.23 | 80.36 | 84.91 | 82.80 | 88.15 | 94.55 |
| Korfball | 46.04 | 59.62 | 69.71 | 57.74 | 67.35 | 75.54 | 65.88 | 72.73 | 79.59 | 69.93 | 78.11 | 87.72 | 75.76 | 85.85 | 99.43 |
| Modern Pentathlon | 43.31 | 55.86 | 65.34 | 55.09 | 63.94 | 71.54 | 63.28 | 69.56 | 75.84 | 67.59 | 75.18 | 84.04 | 73.79 | 83.27 | 95.82 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 48.49 | 56.58 | 63.13 | 59.06 | 64.77 | 69.86 | 66.41 | 70.47 | 74.53 | 71.08 | 76.16 | 81.88 | 77.81 | 84.36 | 92.44 |
| Rowing | 54.76 | 64.18 | 71.69 | 65.98 | 72.64 | 78.51 | 73.79 | 78.52 | 83.26 | 78.53 | 84.40 | 91.06 | 85.35 | 92.86 | 102.28 |
| Rugby | 55.97 | 66.58 | 75.53 | 74.12 | 81.56 | 88.32 | 86.73 | 91.97 | 97.21 | 95.62 | 102.38 | 109.82 | 108.41 | 117.37 | 127.97 |
| Sailing | 46.73 | 57.09 | 65.52 | 61.09 | 68.35 | 74.82 | 71.08 | 76.18 | 81.28 | 77.54 | 84.01 | 91.27 | 86.84 | 95.27 | 105.63 |
| Soccer | 54.43 | 61.14 | 66.68 | 63.90 | 68.67 | 72.96 | 70.48 | 73.90 | 77.33 | 74.85 | 79.14 | 83.91 | 81.13 | 86.67 | 93.37 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 50.65 | 57.91 | 63.93 | 61.18 | 66.33 | 70.96 | 68.50 | 72.18 | 75.85 | 73.39 | 78.02 | 83.17 | 80.42 | 86.44 | 93.70 |
| Tennis | 45.32 | 56.22 | 64.75 | 57.61 | 65.27 | 71.96 | 66.15 | 71.56 | 76.97 | 71.16 | 77.85 | 85.51 | 78.37 | 86.90 | 97.80 |
| Triathlon | 52.60 | 57.34 | 61.26 | 59.16 | 62.55 | 65.60 | 63.73 | 66.17 | 68.62 | 66.74 | 69.79 | 73.18 | 71.08 | 75.00 | 79.74 |
| Volleyball | 65.18 | 76.29 | 84.88 | 76.15 | 84.06 | 90.94 | 83.77 | 89.46 | 95.15 | 87.98 | 94.86 | 102.77 | 94.04 | 102.63 | 113.74 |
| Wrestling and Judo | 50.18 | 56.27 | 61.48 | 60.98 | 65.29 | 69.24 | 68.49 | 71.56 | 74.64 | 73.89 | 77.83 | 82.15 | 81.65 | 86.85 | 92.95 |

NA: data not presented for $\mathrm{n}<8$

Table S2 - Height (cm) percentiles by sport and sex


Table S3 - Body mass index $\left(\mathrm{kg} / \mathrm{m}^{2}\right)$ percentiles by sport and sex


NA: data not presented for $\mathrm{n}<8$.

Table S4 - Sum of seven skinfolds [triceps + subscapular + biceps + suprailiac + abdominal + thigh + medial calf ( mm )] percentiles by sport and sex


Table S5 - Sum of appendicular skinfolds [triceps + biceps + thigh + medial calf (mm)] percentiles by sport and sex

| Sport | Low | 0.05 | High | Low | 0.25 <br> Estimate | High | Low | Median <br> Estimate | High | Low | 0.75Estimate | High | Low | 0.95 | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Estimate |  |  |  |  |  |  |  |  |  |  |  | Estimate |  |
| Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 0.00 | 10.62 | 23.06 | 15.14 | 25.93 | 35.42 | 29.13 | 36.57 | 44.01 | 37.72 | 47.21 | 58.00 | 50.08 | 62.52 | 78.13 |
| Basketball | 18.69 | 34.51 | 47.56 | 42.39 | 53.44 | 63.36 | 58.86 | 66.60 | 74.35 | 69.84 | 79.76 | 90.82 | 85.64 | 98.70 | 114.51 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 10.29 | 28.44 | 42.82 | 30.90 | 43.74 | 55.01 | 45.23 | 54.38 | 63.49 | 53.71 | 65.02 | 77.82 | 65.91 | 80.32 | 98.44 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | 10.52 | 34.51 | 53.26 | 32.37 | 49.76 | 65.00 | 47.55 | 60.36 | 73.17 | 55.71 | 70.96 | 88.35 | 67.45 | 86.21 | 110.19 |
| Modern Pentathlon | 0.84 | 24.51 | 42.91 | 21.80 | 38.94 | 53.93 | 36.37 | 48.98 | 61.59 | 44.02 | 59.01 | 76.16 | 55.04 | 73.44 | 97.11 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 2.97 | 27.67 | 46.92 | 27.06 | 44.71 | 60.12 | 43.80 | 56.55 | 69.30 | 52.97 | 68.39 | 86.04 | 66.18 | 85.43 | 110.13 |
| Rowing | 8.44 | 32.17 | 50.71 | 29.31 | 46.59 | 61.75 | 43.81 | 56.62 | 69.43 | 51.49 | 66.65 | 83.93 | 62.54 | 81.07 | 104.80 |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 15.08 | 31.99 | 45.58 | 35.61 | 47.56 | 58.15 | 49.88 | 58.38 | 66.88 | 58.61 | 69.20 | 81.15 | 71.17 | 84.76 | 101.68 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 11.66 | 26.17 | 37.87 | 30.38 | 40.57 | 49.59 | 43.39 | 50.58 | 57.74 | 51.54 | 60.59 | 70.76 | 63.27 | 74.99 | 89.48 |
| Tennis | 29.68 | 51.05 | 67.95 | 49.43 | 65.05 | 78.84 | 63.15 | 74.78 | 86.41 | 70.72 | 84.51 | 100.13 | 81.61 | 98.51 | 119.88 |
| Triathlon | 0.00 | 18.80 | 39.48 | 14.95 | 34.38 | 51.26 | 30.95 | 45.21 | 59.45 | 39.14 | 56.05 | 75.45 | 50.92 | 71.63 | 98.46 |
| Volleyball | 7.90 | 31.17 | 49.49 | 33.39 | 49.86 | 64.28 | 51.11 | 62.85 | 74.55 | 61.38 | 75.84 | 92.27 | 76.17 | 94.54 | 117.76 |
| Wrestling and Judo | 11.62 | 33.74 | 51.38 | 38.13 | 53.69 | 67.42 | 56.56 | 67.56 | 78.57 | 67.70 | 81.43 | 96.99 | 83.74 | 101.38 | 123.50 |
|  |  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |
| Archery and Shooting | 1.42 | 22.75 | 36.30 | 18.70 | 32.50 | 43.10 | 30.71 | 39.27 | 47.83 | 35.44 | 46.05 | 59.84 | 42.24 | 55.79 | 77.12 |
| Athletics | 4.53 | 11.28 | 16.29 | 12.14 | 16.67 | 20.48 | 17.43 | 20.42 | 23.39 | 20.35 | 24.17 | 28.68 | 24.54 | 29.56 | 36.29 |
| Basketball | 9.05 | 17.23 | 23.82 | 21.54 | 27.09 | 31.98 | 30.22 | 33.94 | 37.66 | 35.90 | 40.79 | 46.34 | 44.06 | 50.65 | 58.83 |
| Fencing | 0.00 | 11.13 | 23.27 | 9.29 | 21.21 | 30.47 | 20.92 | 28.21 | 35.48 | 25.92 | 35.21 | 47.11 | 33.13 | 45.28 | 63.84 |
| Gymnastics | 1.51 | 12.99 | 21.30 | 13.84 | 21.46 | 27.77 | 22.42 | 27.34 | 32.27 | 26.92 | 33.23 | 40.84 | 33.39 | 41.70 | 53.17 |
| Handball | 1.51 | 17.44 | 28.91 | 18.29 | 28.86 | 37.58 | 29.96 | 36.79 | 43.61 | 35.99 | 44.72 | 55.28 | 44.67 | 56.13 | 72.07 |
| Hockey Rink | 2.00 | 13.26 | 22.31 | 19.49 | 27.05 | 33.72 | 31.65 | 36.64 | 41.65 | 39.57 | 46.22 | 53.81 | 50.98 | 60.01 | 71.30 |
| Korfball | 0.00 | 14.31 | 23.79 | 12.39 | 21.80 | 29.14 | 21.17 | 27.02 | 32.85 | 24.88 | 32.23 | 41.63 | 30.22 | 39.73 | 54.25 |
| Modern Pentathlon | 0.00 | 9.60 | 20.51 | 8.74 | 19.18 | 27.52 | 19.27 | 25.83 | 32.40 | 24.14 | 32.49 | 42.92 | 31.15 | 42.06 | 58.07 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 0.00 | 9.13 | 19.44 | 12.65 | 21.70 | 29.37 | 24.61 | 30.44 | 36.27 | 31.51 | 39.18 | 48.23 | 41.44 | 51.76 | 65.44 |
| Rowing | 1.81 | 11.95 | 19.61 | 14.12 | 20.88 | 26.64 | 22.68 | 27.10 | 31.53 | 27.57 | 33.31 | 40.09 | 34.61 | 42.25 | 52.40 |
| Rugby | 0.00 | 2.68 | 17.55 | 16.34 | 28.47 | 39.20 | 38.52 | 46.39 | 54.25 | 53.58 | 64.31 | 76.44 | 75.23 | 90.10 | 108.35 |
| Sailing | 0.00 | 9.32 | 22.39 | 15.96 | 27.15 | 36.77 | 32.31 | 39.54 | 46.77 | 42.30 | 51.93 | 63.11 | 56.68 | 69.76 | 86.62 |
| Soccer | 3.73 | 13.70 | 20.80 | 13.74 | 20.37 | 25.83 | 20.70 | 25.01 | 29.32 | 24.20 | 29.65 | 36.28 | 29.23 | 36.33 | 46.30 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 5.70 | 12.80 | 18.45 | 16.03 | 20.84 | 25.04 | 23.21 | 26.42 | 29.62 | 27.79 | 32.01 | 36.80 | 34.39 | 40.05 | 47.14 |
| Tennis | 9.13 | 19.09 | 26.33 | 19.50 | 26.18 | 31.75 | 26.71 | 31.11 | 35.52 | 30.47 | 36.04 | 42.72 | 35.89 | 43.14 | 53.09 |
| Triathlon | 7.37 | 13.30 | 17.92 | 15.11 | 19.13 | 22.62 | 20.49 | 23.19 | 25.88 | 23.76 | 27.24 | 31.27 | 28.45 | 33.08 | 39.01 |
| Volleyball | 0.00 | 12.97 | 22.29 | 13.11 | 21.83 | 28.94 | 22.38 | 27.98 | 33.57 | 27.01 | 34.14 | 42.84 | 33.66 | 43.00 | 56.19 |
| Wrestling and Judo | 4.22 | 10.78 | 16.19 | 15.74 | 20.19 | 24.16 | 23.75 | 26.73 | 29.70 | 29.29 | 33.27 | 37.70 | 37.25 | 42.68 | 49.22 |

Table S6 - Sum of arm skinfolds [triceps + biceps (mm)] percentiles by sport and sex


Table S7 - Sum of leg skinfolds [thigh + medial calf (mm)] percentiles by sport and sex

| Sport | Low | 0.05 <br> Estimate | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | 0.75Estimate | High | Low | $0.95$ <br> Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 0.00 | 4.46 | 13.24 | 7.31 | 15.00 | 21.61 | 17.21 | 22.32 | 27.43 | 23.03 | 29.64 | 37.32 | 31.39 | 40.17 | 51.56 |
| Basketball | 11.18 | 21.95 | 30.70 | 26.92 | 34.36 | 40.96 | 37.86 | 42.97 | 48.09 | 44.99 | 51.59 | 59.03 | 55.25 | 63.99 | 74.77 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 4.59 | 17.45 | 27.30 | 18.71 | 27.58 | 35.22 | 28.52 | 34.63 | 40.73 | 34.03 | 41.67 | 50.54 | 41.95 | 51.80 | 64.67 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | 3.00 | 19.59 | 31.81 | 17.49 | 29.11 | 38.91 | 27.57 | 35.72 | 43.85 | 32.51 | 42.34 | 53.93 | 39.62 | 51.86 | 68.43 |
| Modern Pentathlon | 0.00 | 13.06 | 25.40 | 10.55 | 22.33 | 32.24 | 20.58 | 28.78 | 36.99 | 25.33 | 35.22 | 47.02 | 32.17 | 44.49 | 61.44 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 0.00 | 16.05 | 28.23 | 14.99 | 26.38 | 36.03 | 25.68 | 33.56 | 41.44 | 31.10 | 40.74 | 52.13 | 38.89 | 51.07 | 67.51 |
| Rowing | 2.94 | 19.02 | 30.86 | 16.50 | 27.81 | 37.37 | 25.93 | 33.92 | 41.90 | 30.46 | 40.02 | 51.33 | 36.97 | 48.81 | 64.89 |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 7.33 | 19.99 | 29.86 | 22.24 | 30.98 | 38.58 | 32.60 | 38.61 | 44.64 | 38.66 | 46.24 | 55.01 | 47.38 | 57.23 | 69.92 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 6.91 | 16.62 | 24.27 | 19.05 | 25.75 | 31.60 | 27.48 | 32.09 | 36.70 | 32.58 | 38.43 | 45.13 | 39.91 | 47.55 | 57.26 |
| Tennis | 13.40 | 28.99 | 40.70 | 27.32 | 38.33 | 47.75 | 37.00 | 44.81 | 52.65 | 41.90 | 51.30 | 62.33 | 48.95 | 60.63 | 76.26 |
| Triathlon | 0.00 | 10.73 | 23.98 | 7.46 | 20.25 | 30.93 | 17.98 | 26.87 | 35.76 | 22.81 | 33.49 | 46.28 | 29.76 | 43.02 | 61.41 |
| Volleyball | 0.00 | 17.22 | 31.15 | 18.33 | 30.99 | 41.80 | 31.93 | 40.57 | 49.20 | 39.34 | 50.14 | 62.80 | 49.99 | 63.92 | 82.37 |
| Wrestling and Judo | 1.20 | 18.48 | 31.78 | 21.33 | 33.16 | 43.36 | 35.32 | 43.36 | 51.41 | 43.36 | 53.56 | 65.40 | 54.94 | 68.24 | 85.52 |
|  |  |  | Males |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | 0.00 | 12.74 | 21.42 | 10.13 | 19.00 | 25.77 | 17.90 | 23.35 | 28.80 | 20.92 | 27.70 | 36.57 | 25.27 | 33.95 | 47.75 |
| Athletics | 0.00 | 4.81 | 8.66 | 5.54 | 9.01 | 11.91 | 9.69 | 11.93 | 14.17 | 11.95 | 14.85 | 18.32 | 15.20 | 19.05 | 24.29 |
| Basketball | 3.77 | 9.16 | 13.50 | 12.03 | 15.67 | 18.88 | 17.77 | 20.19 | 22.62 | 21.51 | 24.72 | 28.36 | 26.89 | 31.23 | 36.62 |
| Fencing | 0.00 | 4.74 | 13.38 | 3.45 | 11.94 | 18.50 | 11.84 | 16.94 | 22.05 | 15.39 | 21.94 | 30.44 | 20.51 | 29.14 | 42.50 |
| Gymnastics | 0.00 | 6.70 | 12.13 | 7.29 | 12.26 | 16.37 | 12.92 | 16.12 | 19.31 | 15.87 | 19.98 | 24.95 | 20.10 | 25.54 | 33.06 |
| Handball | 1.77 | 11.00 | 17.64 | 11.47 | 17.59 | 22.65 | 18.20 | 22.17 | 26.13 | 21.68 | 26.75 | 32.87 | 26.70 | 33.33 | 42.56 |
| Hockey Rink | 0.00 | 6.51 | 12.61 | 10.75 | 15.85 | 20.32 | 19.00 | 22.34 | 25.68 | 24.35 | 28.83 | 33.92 | 32.06 | 38.16 | 45.79 |
| Korfball | 0.00 | 6.48 | 13.40 | 5.11 | 12.00 | 17.30 | 11.64 | 15.83 | 20.01 | 14.35 | 19.66 | 26.54 | 18.25 | 25.18 | 35.94 |
| Modern Pentathlon | 0.00 | 5.07 | 11.79 | 4.53 | 10.97 | 16.10 | 11.06 | 15.08 | 19.11 | 14.06 | 19.19 | 25.63 | 18.38 | 25.10 | 35.02 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 0.00 | 0.50 | 9.32 | 3.63 | 11.35 | 17.83 | 14.04 | 18.89 | 23.74 | 19.95 | 26.43 | 34.16 | 28.46 | 37.28 | 49.14 |
| Rowing | 0.00 | 5.46 | 10.79 | 7.02 | 11.73 | 15.71 | 13.05 | 16.09 | 19.13 | 16.47 | 20.45 | 25.16 | 21.39 | 26.72 | 33.84 |
| Rugby | 0.00 | 0.99 | 10.10 | 9.37 | 16.79 | 23.36 | 22.97 | 27.78 | 32.58 | 32.19 | 38.76 | 46.18 | 45.45 | 54.56 | 65.76 |
| Sailing | 0.00 | 1.29 | 10.93 | 6.28 | 14.51 | 21.54 | 18.46 | 23.69 | 28.92 | 25.83 | 32.87 | 41.09 | 36.44 | 46.08 | 58.61 |
| Soccer | 0.00 | 6.74 | 11.89 | 6.82 | 11.63 | 15.57 | 11.96 | 15.04 | 18.12 | 14.51 | 18.44 | 23.26 | 18.18 | 23.34 | 30.66 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 1.89 | 6.85 | 10.79 | 9.13 | 12.47 | 15.39 | 14.17 | 16.38 | 18.59 | 17.37 | 20.29 | 23.62 | 21.97 | 25.91 | 30.87 |
| Tennis | 2.97 | 9.89 | 14.89 | 10.22 | 14.83 | 18.65 | 15.27 | 18.27 | 21.26 | 17.88 | 21.70 | 26.31 | 21.64 | 26.64 | 33.56 |
| Triathlon | 2.33 | 6.69 | 10.06 | 8.06 | 10.99 | 13.51 | 12.04 | 13.98 | 15.91 | 14.44 | 16.96 | 19.89 | 17.89 | 21.26 | 25.62 |
| Volleyball | 0.00 | 6.03 | 12.96 | 6.19 | 12.67 | 17.92 | 13.18 | 17.28 | 21.37 | 16.63 | 21.89 | 28.36 | 21.59 | 28.52 | 38.42 |
| Wrestling and Judo | 0.95 | 5.40 | 9.07 | 8.79 | 11.80 | 14.48 | 14.25 | 16.24 | 18.24 | 18.01 | 20.69 | 23.69 | 23.42 | 27.09 | 31.54 |

Table S8 - Sum of trunk skinfolds [subscapular + suprailiac + abdominal (mm)] percentiles by sport and sex


Table S9 - Arm circumference (cm) percentiles by sport and sex


Table S10 - Arm muscle circumference ( cm ) percentiles by sport and sex


Table S11 - Thigh circumference (cm) percentiles by sport and sex


Table S12 - Thigh muscle circumference ( cm ) percentiles by sport and sex


Table S13 - Calf circumference (cm) percentiles by sport and sex


Table S14 - Calf muscle circumference ( cm ) percentiles by sport and sex


Table S15 - Abdominal circumference (cm) percentiles by sport and sex

| Sport | Low | 0.05 | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | $0.75$ <br> Estimate | High | Low | 0.95 | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Estimate |  |  |  |  |  |  |  |  |  |  |  | Estimate |  |
|  |  |  |  |  |  | Females |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 59.84 | 64.43 | 68.23 | 65.15 | 68.54 | 71.60 | 68.85 | 71.39 | 73.94 | 71.19 | 74.25 | 77.64 | 74.56 | 78.36 | 82.95 |
| Basketball | 63.58 | 68.29 | 72.26 | 70.12 | 73.56 | 76.69 | 74.67 | 77.22 | 79.76 | 77.75 | 80.88 | 84.31 | 82.17 | 86.15 | 90.86 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 56.81 | 62.74 | 67.59 | 62.98 | 67.36 | 71.31 | 67.26 | 70.57 | 73.89 | 69.84 | 73.79 | 78.17 | 73.56 | 78.41 | 84.34 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | 60.79 | 69.90 | 77.20 | 68.39 | 75.24 | 81.36 | 73.67 | 78.96 | 84.25 | 76.56 | 82.68 | 89.53 | 80.72 | 88.02 | 97.13 |
| Modern Pentathlon | 53.90 | 65.55 | 74.84 | 63.44 | 72.18 | 79.96 | 70.06 | 76.79 | 83.52 | 73.62 | 81.40 | 90.14 | 78.74 | 88.03 | 99.67 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 62.22 | 69.99 | 76.25 | 69.09 | 74.91 | 80.11 | 73.87 | 78.33 | 82.79 | 76.56 | 81.75 | 87.57 | 80.42 | 86.68 | 94.45 |
| Rowing | 58.63 | 68.66 | 76.70 | 66.73 | 74.31 | 81.06 | 72.37 | 78.23 | 84.10 | 75.40 | 82.16 | 89.73 | 79.77 | 87.80 | 97.84 |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 61.01 | 66.41 | 70.86 | 67.04 | 71.02 | 74.61 | 71.23 | 74.22 | 77.21 | 73.83 | 77.42 | 81.40 | 77.58 | 82.03 | 87.44 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 59.67 | 64.95 | 69.33 | 65.93 | 69.81 | 73.31 | 70.28 | 73.18 | 76.08 | 73.05 | 76.56 | 80.43 | 77.04 | 81.41 | 86.69 |
| Tennis | 62.12 | 71.13 | 78.35 | 69.90 | 76.64 | 82.65 | 75.30 | 80.47 | 85.65 | 78.29 | 84.30 | 91.05 | 82.60 | 89.82 | 98.82 |
| Triathlon | 56.07 | 66.63 | 75.13 | 64.30 | 72.30 | 79.49 | 70.03 | 76.25 | 82.52 | 73.06 | 80.19 | 88.25 | 77.42 | 85.87 | 96.48 |
| Volleyball | 61.73 | 69.44 | 75.69 | 69.48 | 75.18 | 80.28 | 74.87 | 79.17 | 83.47 | 78.06 | 83.16 | 88.85 | 82.65 | 88.90 | 96.61 |
| Wrestling and Judo | 57.97 | 64.23 | 69.36 | 64.91 | 69.50 | 73.63 | 69.72 | 73.16 | 76.60 | 72.69 | 76.82 | 81.41 | 76.96 | 82.09 | 88.35 |
|  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | 49.48 | 66.83 | 79.61 | 63.56 | 75.85 | 86.31 | 73.34 | 82.12 | 90.96 | 78.00 | 88.40 | 100.75 | 84.70 | 97.42 | 114.83 |
| Athletics | 62.27 | 68.01 | 72.59 | 68.50 | 72.63 | 76.28 | 72.84 | 75.84 | 78.85 | 75.40 | 79.05 | 83.18 | 79.09 | 83.67 | 89.42 |
| Basketball | 65.70 | 70.70 | 74.90 | 73.01 | 76.60 | 79.86 | 78.10 | 80.70 | 83.31 | 81.55 | 84.80 | 88.40 | 86.51 | 90.70 | 95.71 |
| Fencing | 56.09 | 67.37 | 75.85 | 65.96 | 74.00 | 80.89 | 72.82 | 78.60 | 84.39 | 76.32 | 83.21 | 91.25 | 81.35 | 89.84 | 101.11 |
| Gymnastics | 58.20 | 66.27 | 72.59 | 66.60 | 72.36 | 77.41 | 72.44 | 76.60 | 80.76 | 75.79 | 80.83 | 86.60 | 80.60 | 86.92 | 95.00 |
| Handball | 60.08 | 70.04 | 77.80 | 70.34 | 77.42 | 83.61 | 77.47 | 82.56 | 87.65 | 81.51 | 87.69 | 94.77 | 87.32 | 95.08 | 105.03 |
| Hockey Rink | 65.80 | 70.98 | 75.31 | 73.45 | 77.15 | 80.52 | 78.76 | 81.45 | 84.13 | 82.38 | 85.74 | 89.45 | 87.58 | 91.92 | 97.09 |
| Korfball | 56.28 | 67.66 | 76.19 | 65.92 | 74.05 | 81.02 | 72.61 | 78.50 | 84.38 | 75.97 | 82.94 | 91.08 | 80.80 | 89.34 | 100.71 |
| Modern Pentathlon | 56.11 | 66.00 | 73.53 | 65.21 | 72.26 | 78.34 | 71.53 | 76.61 | 81.69 | 74.88 | 80.97 | 88.02 | 79.70 | 87.23 | 97.11 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 63.15 | 68.67 | 73.13 | 69.59 | 73.56 | 77.10 | 74.07 | 76.97 | 79.86 | 76.83 | 80.37 | 84.34 | 80.80 | 85.26 | 90.79 |
| Rowing | 61.61 | 69.03 | 74.97 | 70.29 | 75.58 | 80.27 | 76.32 | 80.14 | 83.96 | 80.01 | 84.70 | 89.99 | 85.31 | 91.25 | 98.68 |
| Rugby | 63.86 | 71.21 | 77.46 | 76.21 | 81.43 | 86.21 | 84.79 | 88.54 | 92.28 | 90.87 | 95.64 | 100.86 | 99.61 | 105.86 | 113.21 |
| Sailing | 61.38 | 69.02 | 75.26 | 71.63 | 77.05 | 81.90 | 78.75 | 82.64 | 86.52 | 83.37 | 88.22 | 93.64 | 90.01 | 96.25 | 103.89 |
| Soccer | 59.77 | 68.72 | 75.66 | 68.55 | 74.94 | 80.51 | 74.65 | 79.27 | 83.89 | 78.02 | 83.59 | 89.99 | 82.88 | 89.82 | 98.76 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 63.41 | 68.45 | 72.63 | 70.42 | 74.03 | 77.29 | 75.29 | 77.91 | 80.53 | 78.53 | 81.79 | 85.40 | 83.19 | 87.37 | 92.41 |
| Tennis | 61.96 | 69.47 | 75.34 | 69.56 | 74.94 | 79.66 | 74.84 | 78.75 | 82.65 | 77.84 | 82.55 | 87.93 | 82.15 | 88.03 | 95.53 |
| Triathlon | 63.19 | 68.04 | 72.01 | 69.22 | 72.72 | 75.86 | 73.42 | 75.97 | 78.53 | 76.09 | 79.22 | 82.72 | 79.93 | 83.90 | 88.76 |
| Volleyball | 64.47 | 73.69 | 80.83 | 73.51 | 80.09 | 85.83 | 79.79 | 84.54 | 89.30 | 83.26 | 89.00 | 95.58 | 88.26 | 95.40 | 104.62 |
| Wrestling and Judo | 62.97 | 67.21 | 70.79 | 69.98 | 73.04 | 75.80 | 74.86 | 77.10 | 79.28 | 78.35 | 81.15 | 84.16 | 83.35 | 86.99 | 91.18 |

Table S16 - Hip circumference (cm) percentiles by sport and sex

| Sport | Low | 0.05 | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | 0.75 <br> Estimate | High | Low | 0.95Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Estimate |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  | Females |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 80.60 | 84.11 | 87.47 | 85.93 | 88.95 | 91.91 | 89.64 | 92.31 | 94.99 | 92.72 | 95.68 | 98.69 | 97.16 | 100.52 | 104.03 |
| Basketball | 89.25 | 92.40 | 95.41 | 94.78 | 97.44 | 100.04 | 98.62 | 100.94 | 103.26 | 101.83 | 104.44 | 107.10 | 106.46 | 109.47 | 112.62 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 79.56 | 83.60 | 87.48 | 85.06 | 88.58 | 92.04 | 88.89 | 92.05 | 95.21 | 92.06 | 95.51 | 99.03 | 96.62 | 100.50 | 104.54 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | 80.01 | 85.18 | 90.19 | 85.47 | 90.11 | 94.68 | 89.27 | 93.54 | 97.81 | 92.40 | 96.97 | 101.61 | 96.90 | 101.90 | 107.08 |
| Modern Pentathlon | 80.63 | 86.04 | 91.28 | 86.11 | 90.98 | 95.78 | 89.92 | 94.42 | 98.92 | 93.05 | 97.85 | 102.73 | 97.56 | 102.80 | 108.21 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 81.12 | 85.93 | 90.51 | 86.54 | 90.83 | 94.99 | 90.31 | 94.24 | 98.10 | 93.42 | 97.64 | 101.87 | 97.89 | 102.54 | 107.29 |
| Rowing | 82.66 | 88.09 | 93.36 | 88.13 | 93.03 | 97.86 | 91.93 | 96.46 | $\begin{aligned} & 100.9 \\ & 9 \end{aligned}$ | 95.05 | 99.89 | 104.79 | 99.55 | 104.82 | 110.26 |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 83.52 | 87.30 | 90.92 | 89.05 | 92.31 | 95.51 | 92.89 | 95.79 | 98.70 | 96.08 | 99.27 | 102.54 | $\begin{aligned} & 100.6 \\ & 6 \end{aligned}$ | 104.29 | 108.06 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 82.49 | 85.92 | 89.21 | 87.81 | 90.76 | 93.64 | 91.52 | 94.12 | 96.72 | 94.59 | 97.48 | 100.42 | 99.02 | 102.31 | 105.75 |
| Tennis | 84.83 | 89.81 | 94.63 | 90.28 | 94.74 | 99.12 | 94.07 | 98.16 | 102.25 | 97.20 | 101.59 | 106.04 | 101.69 | 106.51 | 111.49 |
| Triathlon | 78.88 | 84.53 | 90.06 | 84.34 | 89.45 | 94.54 | 88.13 | 92.87 | 97.66 | 91.25 | 96.28 | 101.45 | 95.73 | 101.20 | 106.91 |
| Volleyball | 88.24 | 92.62 | 96.76 | 93.86 | 97.71 | 101.42 | 97.77 | 101.24 | 104.65 | 101.00 | 104.78 | 108.56 | 105.66 | 109.87 | 114.18 |
| Wrestling and Judo | 82.11 | 85.89 | 89.52 | 87.55 | 90.82 | 94.03 | 91.33 | 94.25 | 97.17 | 94.47 | 97.68 | 100.96 | 98.99 | 102.62 | 106.40 |
|  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | 79.74 | 87.90 | 94.55 | 86.43 | 92.66 | 98.27 | 91.08 | 95.97 | 100.86 | 93.66 | 99.27 | 105.50 | 97.39 | 104.03 | 112.19 |
| Athletics | 83.42 | 88.31 | 92.37 | 88.88 | 92.52 | 95.82 | 92.68 | 95.45 | 98.21 | 95.08 | 98.37 | 102.01 | 98.52 | 102.58 | 107.47 |
| Basketball | 86.43 | 90.66 | 94.29 | 92.63 | 95.75 | 98.63 | 96.94 | 99.29 | 101.64 | 99.95 | 102.83 | 105.95 | 104.28 | 107.91 | 112.15 |
| Fencing | 78.82 | 87.03 | 93.72 | 86.26 | 92.44 | 98.00 | 91.43 | 96.21 | 100.98 | 94.41 | 99.97 | 106.16 | 98.70 | 105.39 | 113.60 |
| Gymnastics | 78.19 | 84.09 | 88.97 | 84.50 | 88.90 | 92.88 | 88.89 | 92.24 | 95.59 | 91.61 | 95.59 | 99.98 | 95.51 | 100.39 | 106.30 |
| Handball | 84.23 | 90.97 | 96.53 | 91.30 | 96.33 | 100.87 | 96.22 | 100.05 | 103.88 | 99.23 | 103.77 | 108.80 | 103.57 | 109.13 | 115.87 |
| Hockey Rink | 86.10 | 89.93 | 93.21 | 91.76 | 94.58 | 97.18 | 95.70 | 97.82 | 99.94 | 98.46 | 101.06 | 103.88 | 102.43 | 105.71 | 109.54 |
| Korfball | 77.25 | 85.30 | 91.85 | 84.32 | 90.41 | 95.88 | 89.23 | 93.96 | 98.69 | 92.03 | 97.51 | 103.60 | 96.06 | 102.62 | 110.67 |
| Modern Pentathlon | 78.05 | 85.26 | 91.15 | 84.91 | 90.32 | 95.18 | 89.67 | 93.83 | 97.99 | 92.48 | 97.35 | 102.75 | 96.51 | 102.40 | 109.61 |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 83.41 | 87.72 | 91.33 | 88.62 | 91.81 | 94.73 | 92.23 | 94.66 | 97.09 | 94.59 | 97.51 | 100.71 | 97.99 | 101.60 | 105.91 |
| Rowing | 82.97 | 88.29 | 92.74 | 89.28 | 93.23 | 96.82 | 93.66 | 96.66 | 99.65 | 96.50 | 100.09 | 104.04 | 100.57 | 105.02 | 110.35 |
| Rugby | 86.08 | 91.45 | 96.08 | 95.11 | 99.01 | 102.60 | 101.38 | 104.26 | 107.14 | 105.91 | 109.51 | 113.41 | 112.44 | 117.07 | 122.43 |
| Sailing | 81.73 | 87.05 | 91.55 | 88.91 | 92.82 | 96.40 | 93.90 | 96.83 | 99.77 | 97.26 | 100.84 | 104.76 | 102.11 | 106.61 | 111.94 |
| Soccer | 80.63 | 87.25 | 92.70 | 87.31 | 92.27 | 96.74 | 91.96 | 95.75 | 99.55 | 94.76 | 99.24 | 104.19 | 98.80 | 104.25 | 110.87 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 82.85 | 86.74 | 90.06 | 88.29 | 91.16 | 93.80 | 92.06 | 94.23 | 96.40 | 94.66 | 97.30 | 100.17 | 98.40 | 101.72 | 105.60 |
| Tennis | 80.79 | 86.40 | 91.04 | 86.61 | 90.81 | 94.61 | 90.66 | 93.88 | 97.10 | 93.14 | 96.95 | 101.14 | 96.72 | 101.36 | 106.96 |
| Triathlon | 77.69 | 82.40 | 86.37 | 83.72 | 87.20 | 90.37 | 87.91 | 90.54 | 93.16 | 90.70 | 93.87 | 97.35 | 94.71 | 98.68 | 103.39 |
| Volleyball | 90.64 | 96.01 | 100.45 | 95.98 | 100.03 | 103.68 | 99.70 | 102.82 | 105.93 | 101.95 | 105.61 | 109.65 | 105.19 | 109.62 | 115.00 |
| Wrestling and Judo | 81.70 | 85.15 | 88.16 | 87.44 | 89.98 | 92.33 | 91.43 | 93.33 | 95.23 | 94.33 | 96.68 | 99.22 | 98.51 | 101.51 | 104.96 |

Table S17 - Whole-body bone mineral content (g) percentiles by sport and sex


Table S18 - Whole-body bone mineral density (g/cm3) percentiles by sport and sex


Table S19 - Whole-body fat mass (kg) percentiles by sport and sex


Table S2O - Whole-body fat mass index $\left(\mathrm{kg} / \mathrm{m}^{2}\right)$ percentiles by sport and sex


Table S21 - Whole body fat mass (\%) percentiles by sport and sex


Table S22 - Whole-body fat-free mass (kg) percentiles by sport and sex


Table S23 - Whole-body fat-free mass index $\left(\mathrm{kg} / \mathrm{m}^{2}\right)$ percentiles by sport and sex


Table S24 - Whole-body lean soft tissue (kg) percentiles by sport and sex

| Sport | Low | $0.05$ <br> Estimate | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | $0.75$ <br> Estimate | High | Low | 0.95 <br> Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  |  |  |  |  |  | Females |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 35.72 | 38.31 | 40.90 | 40.71 | 43.27 | 45.83 | 44.19 | 46.73 | 49.26 | 47.62 | 50.18 | 52.74 | 52.55 | 55.14 | 57.73 |
| Basketball | 35.29 | 37.08 | 38.88 | 40.29 | 42.05 | 43.81 | 43.76 | 45.50 | 47.24 | 47.19 | 48.95 | 50.72 | 52.12 | 53.92 | 55.71 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 24.67 | 27.64 | 30.64 | 29.67 | 32.61 | 35.58 | 33.14 | 36.06 | 39.01 | 36.57 | 39.51 | 42.48 | 41.51 | 44.48 | 47.48 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 32.03 | 34.26 | 36.49 | 37.03 | 39.22 | 41.42 | 40.50 | 42.68 | 44.85 | 43.93 | 46.13 | 48.32 | 48.87 | 51.09 | 53.32 |
| Tennis | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Triathlon | 29.83 | 32.84 | 36.37 | 34.83 | 37.81 | 41.31 | 38.30 | 41.26 | 44.74 | 41.73 | 44.71 | 48.21 | 46.67 | 49.68 | 53.21 |
| Volleyball | 35.03 | 37.65 | 40.23 | 40.03 | 42.61 | 45.17 | 43.50 | 46.06 | 48.60 | 46.93 | 49.52 | 52.07 | 51.87 | 54.48 | 57.07 |
| Wrestling and Judo | 29.96 | 32.65 | 35.32 | 34.96 | 37.62 | 40.25 | 38.43 | 41.07 | 43.68 | 41.86 | 44.52 | 47.16 | 46.80 | 49.49 | 52.16 |
|  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 42.32 | 51.34 | 58.23 | 50.12 | 56.63 | 62.26 | 55.54 | 60.30 | 65.06 | 58.35 | 63.98 | 70.48 | 62.38 | 69.26 | 78.28 |
| Basketball | 46.56 | 52.63 | 57.69 | 55.60 | 59.92 | 63.82 | 61.89 | 64.98 | 68.08 | 66.14 | 70.05 | 74.36 | 72.27 | 77.33 | 83.40 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Handball | 46.37 | 53.24 | 58.88 | 55.60 | 60.50 | 64.88 | 62.03 | 65.54 | 69.06 | 66.20 | 70.58 | 75.48 | 72.20 | 77.84 | 84.71 |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 36.02 | 45.98 | 53.61 | 45.27 | 52.39 | 58.54 | 51.70 | 56.84 | 61.97 | 55.13 | 61.29 | 68.40 | 60.06 | 67.69 | 77.65 |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | 43.33 | 51.34 | 57.93 | 54.54 | 60.22 | 65.31 | 62.34 | 66.39 | 70.44 | 67.47 | 72.56 | 78.23 | 74.85 | 81.44 | 89.45 |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 44.82 | 50.75 | 55.53 | 51.98 | 56.21 | 59.98 | 56.95 | 60.01 | 63.07 | 60.04 | 63.80 | 68.04 | 64.48 | 69.27 | 75.19 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 40.76 | 47.03 | 52.17 | 49.19 | 53.64 | 57.64 | 55.04 | 58.24 | 61.44 | 58.85 | 62.84 | 67.30 | 64.32 | 69.46 | 75.72 |
| Tennis | 33.38 | 45.01 | 53.83 | 43.65 | 51.96 | 59.11 | 50.79 | 56.78 | 62.78 | 54.46 | 61.61 | 69.92 | 59.74 | 68.55 | 80.18 |
| Triathlon | 43.39 | 47.50 | 50.90 | 48.96 | 51.91 | 54.56 | 52.82 | 54.97 | 57.11 | 55.37 | 58.03 | 60.98 | 59.03 | 62.43 | 66.54 |
| Volleyball | 50.12 | 59.89 | 67.52 | 59.98 | 66.97 | 73.08 | 66.84 | 71.89 | 76.94 | 70.70 | 76.81 | 83.80 | 76.26 | 83.89 | 93.66 |
| Wrestling and Judo | 40.13 | 46.08 | 51.04 | 48.92 | 53.16 | 56.98 | 55.04 | 58.07 | 61.11 | 59.16 | 62.99 | 67.22 | 65.10 | 70.06 | 76.02 |

Table S25-Subtotal* bone mineral content (g) percentiles by sport and sex

| Sport | Low | 0.05 |  | 0.25 |  | High | Low | Median <br> Estimate | High | Low | 0.75 | High | Low | 0.95 | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Estimate | High | Low | Estimate |  |  |  |  |  | Estimate |  |  | Estimate |  |
| Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 1324 | 1844 | 2258 | 1810 | 2191 | 2529 | 2148 | 2432 | 2718 | 2336 | 2673 | 3056 | 2608 | 3020 | 3542 |
| Basketball | 1655 | 2003 | 2297 | 2195 | 2443 | 2669 | 2571 | 2749 | 2928 | 2829 | 3055 | 3303 | 3202 | 3495 | 3843 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Handball | 1723 | 2075 | 2367 | 2218 | 2469 | 2696 | 2561 | 2743 | 2925 | 2790 | 3017 | 3269 | 3119 | 3412 | 3763 |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 1145 | 1646 | 2043 | 1640 | 2004 | 2325 | 1984 | 2253 | 2521 | 2180 | 2503 | 2866 | 2463 | 2861 | 3361 |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | 1831 | 2143 | 2405 | 2281 | 2506 | 2709 | 2595 | 2757 | 2920 | 2806 | 3009 | 3233 | 3110 | 3371 | 3684 |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 1604 | 1913 | 2167 | 1996 | 2218 | 2419 | 2268 | 2431 | 2593 | 2443 | 2643 | 2866 | 2694 | 2948 | 3257 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 1106 | 1434 | 1706 | 1572 | 1805 | 2015 | 1896 | 2063 | 2229 | 2110 | 2320 | 2553 | 2420 | 2691 | 3020 |
| Tennis | 1050 | 1534 | 1917 | 1504 | 1858 | 2170 | 1820 | 2083 | 2346 | 1996 | 2309 | 2663 | 2250 | 2633 | 3117 |
| Triathlon | 1232 | 1457 | 1645 | 1554 | 1715 | 1861 | 1778 | 1895 | 2011 | 1928 | 2074 | 2235 | 2144 | 2332 | 2557 |
| Volleyball | 1911 | 2432 | 2851 | 2468 | 2846 | 3182 | 2855 | 3133 | 3412 | 3085 | 3421 | 3799 | 3416 | 3835 | 4355 |
| Wrestling and Judo | 1551 | 1830 | 2066 | 1978 | 2178 | 2360 | 2276 | 2420 | 2564 | 2480 | 2662 | 2862 | 2774 | 3010 | 3290 |
|  |  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 873 | 1344 | 1706 | 1355 | 1685 | 1972 | 1690 | 1923 | 2157 | 1875 | 2161 | 2492 | 2142 | 2502 | 2974 |
| Basketball | 905 | 1314 | 1645 | 1469 | 1753 | 2006 | 1861 | 2059 | 2257 | 2112 | 2364 | 2649 | 2473 | 2803 | 3213 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 584 | 1036 | 1376 | 1005 | 1324 | 1595 | 1299 | 1523 | 1747 | 1451 | 1723 | 2040 | 1670 | 2011 | 2462 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 853 | 1146.1 | 1377 | 1187 | 1394 | 1575 | 1419 | 1566 | 1712 | 1556 | 1738 | 1944 | 1754 | 1985 | 2279 |
| Tennis | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Triathlon | 629 | 1018 | 1310 | 969 | 1245 | 1482 | 1206 | 1403 | 1601 | 1325 | 1561 | 1838 | 1496 | 1788 | 2178 |
| Volleyball | 928 | 1373 | 1716 | 1383 | 1696 | 1968 | 1699 | 1921 | 2143 | 1874 | 2145 | 2459 | 2125 | 2468 | 2914 |
| Wrestling and Judo | 796 | 1255 | 1605 | 1255 | 1578 | 1856 | 1575 | 1802 | 2030 | 1749 | 2027 | 2349 | 1999 | 2349 | 2808 |

'Whole-body minus the head

Table S26 - Subtotal* fat mass (kg) percentiles by sport and sex


Whole-body minus the head

Table S27-Subtotal* fat mass (\%) percentiles by sport and sex

"Whole-body minus the head

Table S28-Subtotal* fat-free mass (kg) percentiles by sport and sex


Whole-body minus the head

Table S29 - Subtotal* lean soft tissue (kg) percentiles by sport and sex

"Whole-body minus the head

Table S30 - Appendicular* bone mineral content (g) percentiles by sport and sex

| Sport | Low | $0.05$ <br> Estimate | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | 0.75Estimate | High | Low | $0.95$ <br> Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 625 | 924 | 1153 | 929 | 1139 | 1320 | 1139 | 1287 | 1435 | 1255 | 1436 | 1646 | 1421 | 1650 | 1950 |
| Basketball | 613 | 875 | 1087 | 972 | 1154 | 1316 | 1221 | 1348 | 1475 | 1381 | 1542 | 1725 | 1610 | 1822 | 2084 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 307 | 648 | 901 | 625 | 862 | 1064 | 846 | 1012 | 1177 | 959 | 1161 | 1398 | 1122 | 1376 | 1717 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 467 | 691 | 865 | 722 | 879 | 1015 | 900 | 1009 | 1118 | 1004 | 1140 | 1296 | 1153 | 1328 | 1551 |
| Tennis | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Triathlon | 328 | 665 | 913 | 623 | 859 | 1059 | 828 | 994 | 1160 | 929 | 1129 | 1365 | 1075 | 1324 | 1660 |
| Volleyball | 611 | 911 | 1140 | 915 | 1125 | 1307 | 1126 | 1274 | 1423 | 1242 | 1424 | 1634 | 1409 | 1638 | 1938 |
| Wrestling and Judo | 503 | 811 | 1045 | 810 | 1026 | 1211 | 1023 | 1175 | 1326 | 1139 | 1324 | 1540 | 1305 | 1539 | 1847 |
|  |  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 718 | 1116 | 1430 | 1092 | 1382 | 1637 | 1352 | 1567 | 1781 | 1496 | 1752 | 2041 | 1704 | 2018 | 2415 |
| Basketball | 997 | 1246 | 1456 | 1386 | 1563 | 1724 | 1657 | 1783 | 1910 | 1843 | 2003 | 2180 | 2110 | 2320 | 2570 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Handball | 1038 | 1289 | 1498 | 1393 | 1572 | 1733 | 1639 | 1768 | 1897 | 1803 | 1964 | 2143 | 2038 | 2247 | 2498 |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 604 | 972 | 1263 | 971 | 1237 | 1471 | 1226 | 1421 | 1615 | 1370 | 1605 | 1870 | 1578 | 1869 | 2237 |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | 1128 | 1361 | 1555 | 1465 | 1632 | 1781 | 1699 | 1820 | 1939 | 1857 | 2008 | 2173 | 2083 | 2278 | 2510 |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 943 | 1166 | 1349 | 1227 | 1388 | 1532 | 1425 | 1542 | 1659 | 1552 | 1696 | 1857 | 1734 | 1918 | 2141 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 680 | 904 | 1091 | 1000 | 1159 | 1303 | 1222 | 1336 | 1450 | 1369 | 1512 | 1672 | 1581 | 1767 | 1992 |
| Tennis | 571 | 936 | 1223 | 915 | 1181 | 1414 | 1155 | 1351 | 1547 | 1288 | 1521 | 1786 | 1478 | 1766 | 2131 |
| Triathlon | 800 | 958 | 1091 | 1027 | 1140 | 1243 | 1184 | 1266 | 1349 | 1290 | 1393 | 1506 | 1442 | 1574 | 1733 |
| Volleyball | 1075 | 1478 | 1801 | 1510 | 1800 | 2058 | 1812 | 2024 | 2237 | 1991 | 2248 | 2539 | 2248 | 2571 | 2974 |
| Wrestling and Judo | 926 | 1120 | 1282 | 1223 | 1361 | 1486 | 1429 | 1529 | 1628 | 1571 | 1697 | 1835 | 1775 | 1938 | 2132 |

Table S31 - Appendicular* fat-mass (kg) percentiles by sport and sex

*(right arm + left arm + right leg + left leg)

Table S32 - Appendicular* fat mass (\%) percentiles by sport and sex

| Sport | Low | 0.05Estimate | High | Low | $0.25$ <br> Estimate | High | Low | Median <br> Estimate | High | Low | 0.75 <br> Estimate | High | Low | 0.95Estimate | High |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
|  | Females |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 3.44 | 12.09 | 18.37 | 12.13 | 17.95 | 22.82 | 18.16 | 22.03 | 25.91 | 21.25 | 26.10 | 31.95 | 25.70 | 31.97 | 40.63 |
| Basketball | 15.53 | 21.49 | 26.24 | 23.50 | 27.62 | 31.24 | 29.04 | 31.88 | 34.72 | 32.52 | 36.14 | 40.26 | 37.52 | 42.27 | 48.23 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | 11.89 | 21.06 | 27.61 | 20.07 | 26.34 | 31.52 | 25.75 | 30.00 | 34.25 | 28.47 | 33.66 | 39.93 | 32.39 | 38.94 | 48.11 |
| Handball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 16.28 | 21.63 | 25.73 | 22.17 | 25.87 | 29.07 | 26.26 | 28.82 | 31.39 | 28.58 | 31.77 | 35.48 | 31.91 | 36.01 | 41.36 |
| Tennis | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Triathlon | 0.00 | 11.59 | 20.96 | 9.76 | 18.93 | 26.23 | 18.16 | 24.04 | 29.90 | 21.82 | 29.14 | 38.30 | 27.10 | 36.48 | 50.38 |
| Volleyball | 12.41 | 20.98 | 27.25 | 20.86 | 26.72 | 31.63 | 26.74 | 30.72 | 34.68 | 29.78 | 34.71 | 40.55 | 34.16 | 40.46 | 49.00 |
| Wrestling and Judo | 5.97 | 17.20 | 25.32 | 16.95 | 24.54 | 30.86 | 24.58 | 29.65 | 34.72 | 28.44 | 34.76 | 42.35 | 33.98 | 42.11 | 53.33 |
|  |  |  |  |  |  |  | Males |  |  |  |  |  |  |  |  |
| Archery and Shooting | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Athletics | 0.00 | 4.46 | 9.03 | 3.79 | 8.18 | 11.74 | 7.89 | 10.76 | 13.63 | 9.78 | 13.34 | 17.73 | 12.49 | 17.05 | 23.64 |
| Basketball | 5.84 | 9.25 | 12.02 | 11.04 | 13.37 | 15.45 | 14.65 | 16.24 | 17.84 | 17.03 | 19.11 | 21.45 | 20.46 | 23.24 | 26.65 |
| Fencing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Gymnastics | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Handball | 3.05 | 7.91 | 11.76 | 9.78 | 13.08 | 15.97 | 14.46 | 16.68 | 18.90 | 17.39 | 20.28 | 23.58 | 21.61 | 25.45 | 30.31 |
| Hockey Rink | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Korfball | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Modern Pentathlon | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Motorsport | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Other combat sports | 1.29 | 7.36 | 11.69 | 7.00 | 11.09 | 14.47 | 10.96 | 13.68 | 16.40 | 12.89 | 16.27 | 20.36 | 15.67 | 20.00 | 26.07 |
| Rowing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Rugby | 3.46 | 9.32 | 14.01 | 11.88 | 15.86 | 19.36 | 17.73 | 20.40 | 23.08 | 21.45 | 24.94 | 28.94 | 26.80 | 31.48 | 37.36 |
| Sailing | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Soccer | 2.13 | 5.98 | 8.96 | 6.95 | 9.56 | 11.82 | 10.30 | 12.05 | 13.80 | 12.28 | 14.53 | 17.15 | 15.14 | 18.11 | 21.97 |
| Surf | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA | NA |
| Swimming | 4.00 | 7.53 | 10.34 | 8.87 | 11.28 | 13.39 | 12.25 | 13.88 | 15.52 | 14.38 | 16.49 | 18.90 | 17.43 | 20.23 | 23.76 |
| Tennis | 0.72 | 9.49 | 15.63 | 8.52 | 14.42 | 19.26 | 13.94 | 17.86 | 21.78 | 16.46 | 21.29 | 27.20 | 20.08 | 26.23 | 35.00 |
| Triathlon | 4.00 | 6.83 | 9.10 | 7.98 | 9.93 | 11.64 | 10.75 | 12.08 | 13.40 | 12.51 | 14.23 | 16.17 | 15.05 | 17.32 | 20.15 |
| Volleyball | 2.43 | 7.84 | 11.83 | 8.01 | 11.68 | 14.76 | 11.89 | 14.35 | 16.80 | 13.93 | 17.01 | 20.68 | 16.86 | 20.85 | 26.27 |
| Wrestling and Judo | 3.71 | 6.90 | 9.47 | 8.54 | 10.71 | 12.64 | 11.89 | 13.36 | 14.84 | 14.09 | 16.01 | 18.19 | 17.25 | 19.83 | 23.01 |

Table S33 - Appendicular* fat-free mass (kg) percentiles by sport and sex

*(right arm + left arm + right leg + left leg)

Table S34 - Appendicular* lean soft tissue (kg) percentiles by sport and sex

*(right arm + left arm + right leg + left leg)

Table S35 - Appendicular* lean soft tissue index $\left(\mathrm{kg} / \mathrm{m}^{2}\right)$ percentiles by sport and sex

*(right arm + left arm + right leg + left leg)

Table S36 - Trunk bone mineral content (g) percentiles by sport and sex


Table S37-Trunk fat mass (kg) percentiles by sport and sex


Table S38 - Trunk fat mass (\%) percentiles by sport and sex


Table S39 - Trunk fat-free mass (kg) percentiles by sport and sex


Table S40 - Trunk lean soft tissue (kg) percentiles by sport and sex



[^0]:    ${ }^{1}$ Santos DA, Gobbo LA, Matias CM, Petroski EL, Gonçalves EM, Cyrino ES, Minderico CS, Sardinha, LB, Silva AM (2013). Body composition in taller individuals using DXA: A validation study for athletic and non-athletic populations. Journal of Sports Sciences. 31(4): 405-13. DOI:
    10.1080/02640414.2012.734918.

[^1]:    ${ }^{2}$ Santos DA, Silva AM, Matias CN, Magalhães JP, Minderico CM, Ekelund U, Sardinha LB (in press) Validity of a combined heart rate and motion sensor for the measurement of free-living energy expenditure in very active individuals. Journal of Science and Medicine in Sports.

[^2]:    ${ }^{3}$ Santos DA, Silva AM, Matias CM, Rocha PM, Alison DB, Sardinha LB (in press). Association of an entire season with body composition in elite junior basketball players. The Journal of Sports Medicine and Physical Fitness.

[^3]:    ${ }^{4}$ Santos DA, Dawson JA, Matias CN, Rocha PM, Minderico CS, Allison DB, Sardinha LB, Silva AM. (in review). Dual Energy X-Ray Absorptiometry and Anthropometry Reference Values for Athletes. Medicine and Science in Sports and Exercise.

[^4]:    ${ }^{a}$ Athletics: includes; ${ }^{\text {b }}$ other combat sports: includes karate, taekwondo, and kickboxing)
    Abbreviations: F, female; M, male; DXA, dual energy X-ray absorptiometry; NA, data not available

[^5]:    Abbreviations: F, female; M, male; BMI, body mass index; $\sum 7$ SKF, sum of seven skinfolds (triceps, subscapular, biceps, suprailiac, abdominal, thigh, and medial calf); MC, muscle circumference.
    *MC: Arm muscle circumference $=$ arm circumference - ( $\pi$ triceps skinfold); thigh muscle circumference $=$ thigh circumference $-(\pi$ thigh skinfold); Calf muscle circumference $=$ medial calf circumference - ( $\pi$ medial skinfold). NA: data not available sports for males ( $p<0.05$ )

[^6]:    Abbreviations: WB, whole body; BMC, bone mineral content; FM, fat mass; FFM, fat-free mass; LST, lean soft tissue; ALST, Appendicular lean soft tissue. "Percentiles were not presented for $n<8$. NA: data not presented for $n<2$.

