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Review paper

BODY COMPOSITION ANALYSIS: THE MOST COMMON TEST MODELS AND RESEARCH METHODS[1]

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Abstract: Accurate and valid analysis of body composition is important for the diagnosis of nutritional status, training impact assessment, the body's response to nutritional and therapeutic interventions, growth and development of the organism, and health risk assessment. With the increasing prevalence of chronic non-communicable diseases, obesity, the need for different models and methods of body composition analysis has also increased. This review paper aimed to show the most commonly used methods for body composition analysis and to give a brief insight into the current techniques. The methods for analysing body composition vary in their accuracy and reliability and in practice the use of appropriate methods depends on the interest and the required accuracy, e.g. dual-energy X-ray absoptiometry (DXA) as the gold standard for determining bone mineral density, or magnetic resonance imaging (MRI) as the gold standard for soft tissue and organ analysis. These methods, along with computed tomography (CT), hydrodensitometry and plethysmography, are routinely used in clinical medicine, and due to their complexity, availability and high prices, they are mostly used in sports medicine for research purposes. In sports medicine, the anthropometric method and bioelectrical impedance analysis (BIA) are mostly used, due to their simplicity and comfort for the examinees. The best assessment of body composition can be obtained by a combination of several methods - a multi-component model, which can increase the accuracy and reliability of the obtained data, and health risk can be assessed with greater certainty.

Key words: *body composition, nutritional status, anthropometry, DXA, BIA, sports medicine*

INTRODUCTION

Body composition analysis (BCA) falls into the domain of interest of sports experts, nutritionists and health professionals. Accurate and valid assessment of nutritional status, morphological and functional capacities has a significant role in assessing the impact of training and achieving top results of an athlete, it enables monitoring the body's response to nutritional and therapeutic interventions, growth and development, and intrinsic and extrinsic factors at different biological levels. With the increasing prevalence of chronic non-communicable diseases, obesity, the need for different models and methods of body composition analysis has also increased, in order to objectively assess the nutritional status, functional capacity, health risk and morbidity. Primarily, the interest in body composition analysis arose so as to determine body fat mass (FM). Technological development and better understanding of body composition analysis and impact on health led to new opportunities to improve current practice.

In addition to the above, body composition analysis also provides information to help identify and assess cachexia, sarcopenia, and obesity. Cachexia is a condition characterized by a loss of adipose tissue and muscle

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mass, which is the result of a number of conditions, and is most often caused by advanced aging or malignant diseases. Sarcopenia is a loss of muscle mass caused by aging, while obesity is a chronic disease, characterized by excessive accumulation of body fat, which leads to numerous health complications: hypertension, type 2 diabetes mellitus, metabolic syndrome, heart and blood vessel diseases, spine and joint diseases, mental disorders (Bhatt, 2016; Rodríguez-Hernández et al., 2013). Obesity is one of the biggest health problems in the world and can be said to take the form of a pandemic. In addition to adults, it is concerning that it is increasingly common in children. All of this is associated with decreased functional abilities, increased morbidity, and mortality (Francis et al., 2017; Peterson & Braunschweig, 2016). The World Health Organization (WHO) data indicates that in 2014, there were 600 million adults and 42 million children under the age of 5 who were overweight (WHO, 2016). This tells us how important it is to identify and use valid methods and models in body composition analysis.

MODELS AND METHODS OF BODY COMPOSITION ANALYSIS

1. Models

Body composition analysis can be done by direct and indirect measurement. Despite the fact that direct measurement is the most accurate method, since it requires tissue dissection, it is not an option for use. There are different models and techniques for indirect measurement.

The most widely used and basic model for body composition analysis is the two-component model (2C), where the body is divided into two parts, fat (FM) and fat-free mass (FFM). The 2C model is based on determining body density and the most commonly used technique is hydrodensitometry. In addition to the 2C model, there is a three-component model (3C), where FFM is divided into two parts, the water content and solids, which consists predominantly of proteins and minerals. The next model, which theoretically gives more valid results than the 3C model, is the four-component (4C) model, which provides body analysis results for fats, minerals, total body water (TBW), and proteins. In addition to the results obtained by hydrodensitometry, it is necessary to additionally use dual-energy X-ray absorptiometry (DXA) to obtain accurate data on bone mineral values, and neutron activation analysis to obtain accurate protein data. Based on the above, it is clear that additional measurements expand the number of tested components in BCA (e.g. the amount of calcium, phosphorus, nitrogen, determining extracellular water) and that it is necessary that the measurement is entirely based on independent results for the given component, to reach the multi-component model (MCM) of body analysis, which provides a large amount of data but requires a combination of different measurement methods (Ellis, 2000; Kuriyan, 2018).

Following the development of the model over time, Wang et al. have combined and presented a model of five levels of body composition, which has become the standard in the BCA research (Wang et al., 1992).

1.1 Five levels of body composition

Current body composition analysis models relate to five levels of body composition. The first one is the atomic level, which contains the main chemical elements (oxygen, carbon, hydrogen, nitrogen, calcium and phosphorus), and whose analysis can provide very important information, such as nitrogen content, which tells us about the total amount of protein in the body, and is determined by neutron activation analysis. The second level consists of the main molecular parts and that is the molecular level, which includes water, proteins, carbohydrates, fats and minerals in bones and soft tissues. For example, essential fats are very important for the cell membrane function. While water and minerals in bones can be determined directly (isotope dilution, DXA), the rest is assessed indirectly. The third level is cellular, which consists of cell mass (fat and fat-free cell mass, where metabolic processes take place), extracellular fluid and extracellular matrix. Attempts have been made to develop certain models, based on anthropometry, for estimating cell mass, but none are in widespread use. The fourth is the tissue-organic level, which consists of tissues, organs and systems. For example, adipose tissue's location is subcutaneous and internal or visceral. It is assessed by indirect methods (e.g. ultrasound, CT). The last level represents the level of the whole body, i.e. the organism as a whole, which is divided into different parts and where it is necessary to use different models and methods of body composition analysis – the multi-component model (WHO Expert Committee, 1995).

2. Methods

2.1 Anthropometric measurements

Anthropometry is a widely used method of measuring body mass, longitudinal skeletal dimensionality, transverse skeletal dimensionality, circumference and thickness of skin folds (SF), especially in the child population. Anthropometric measurements can simply help determine the development of the body, morphological characteristics, nutritional status, the relationship between FM and FFM, the impact of training and nutrition on the body, nutritional interventions, and health risks.

As early as in 1921, an equation was developed for estimating body fat based on anthropometric measurements, by measuring the thickness of skin folds (Matiegka, 1921). By measuring the thickness of the skin folds with a calliper, measures are obtained by which, through the use of equations, FM and FFM are estimated. Based on the most commonly used equations, it is mostly measured at the following ten points: triceps, subscapular, abdominal, suprailiac, thigh, biceps, calf, chest, umbilicus, thorax (J. Wang et al., 2000). In order to obtain precise measurements, it is necessary to use a well-calibrated calliper (e.g. Lange, Holtain) and, as mentioned, well-trained personnel to conduct the procedure in line with the standardized methodology. Special attention should be paid to precisely locating the points, pulling of the skin and placing the instrument at an angle of 90°. The most commonly used equations for determining FM and FFM based on the measures obtained by the SF measurement are Jackson and Pollock (Jackson & Pollock, 1978; Jackson et al., 1980) and Durnin and Vomersley (Durnin & Vomersley, 1974).

Using anthropometric procedures, as mentioned, it is also possible to determine nutritional status by calculating body mass index (BMI), and this is the most commonly used measure for determining overweight and obesity. The examinees' body weight (BW) and body height (BH) are measured, and then the BMI is calculated based on a mathematical formula:

$BMI = BW (kg) / BH (m^2)$

Based on the classification made by the World Health Organization (WHO), a person's nutritional status and health risk are determined (Table 1).

Table 1. *WHO BMI classification*

Classification	BMI (kg/m²)	Risk of comorbidities
Underweight	<18,5	Low (other health risks)
Normal range	18,5 – 24,9	Average
Overweight	>25	
Pre-obese	25,0 – 29,9	Increased
Obese class I	30,0 - 34 ,9	Moderate
Obese class II	35,0 – 39,9	Severe
Obese class III	> 40,0	Very severe

This applies to adults, while percentile tables are used for children and adolescents (WHO, n.d.). This method is not valid when it comes to athletes. Athletes have more muscle mass and a lower percentage of fat, so wrong results will be obtained (Weber et al., 2013). This method does not differentiate between weight gain based on an increase in muscle mass, or that based on fat increase (Taşçilar et al., 2011).

Waist circumference can also be used to assess FM and health effects. Measurements are most often performed in the middle of the upper arm, the middle of the thigh, the waist and the hips. When measuring, attention must be paid to the exact location of the measuring points, the position of the measuring tape (90º in relation to the longitudinal axis of the trunk), that the tape is in contact with the skin, but without pressure and that the reading is made in millimetres (Lohman & Roche, 1988).

Anthropometric procedures seem relatively simple, but still require a well-trained person, in order for the results to be as valid as possible. They are cheap, comfortable for the participants and can be performed on-field.

2.2 Hydrodensitometry (hydrostatic/underwater weighing)

This method is also called densitometry and is based on Archimedes' principles of buoyancy. It represents the 2C model and one of the most accurate methods in the analysis of body composition. It is conducted by immersing the entire body in water. The subject should exhale all air when completely submerged in water. It is based on the measurement of water that the body displaces and the correction of the residual volume, in order to obtain the volume of the body and to calculate the density of the body and body fat percentage (% BF). In people with higher body density, the % BF is lower and vice versa. It is not possible to have insight into adipose tissue distribution (Borga et al., 2018). A person with higher %BF will be lighter in water than a person with higher muscle mass and lower %BF. The density of FM (0.9007g / cm3) is lower than the density of water, while the FFM (1.1 g / cm3) is higher (Brožek et al., 1963). The method itself requires a trained person, special equipment, it is expensive, takes a long time and is not comfortable for the participants. The disadvantages of this method are related to estimates of body volume and residual volume (Buskirk, 1961; Frank, 1969).

2.3 Air displacement plethysmography (ADP)

ADP is a method similar to hydrodensitometry, which it has begun to replace in recent years. It represents a 2C model. The measurement is performed in a closed chamber where the participant sits, which is separated by a membrane (diaphragm) from the reference chamber, which is located on the back. When the subject enters the chamber, there is a change in the air volume, which can be determined by changing the pressure and measuring body volume, so based on body weight and body volume, data on body density can be obtained, including FM and FFM. The distribution of fat and muscle mass cannot be obtained. The method is comfortable, fast, simple, but expensive. It is known as BOD POD, which is a commercial product (COSMED, Concord, California, USA). The results may vary depending on the hydration of the FFM, and therefore the FM and FFM may be underestimated or overestimated (Delisle-Houde et al., 2019; Kendall et al., 2017). The accuracy of hydrodensitometry and ADP is similar, compared to the 4C method (Fields et al., 2001; Millard-Stafford et al., 2001).

2.4 Dual-energy X-ray absorptiometry (DXA)

DXA is based on the transmission of ionizing rays of different energies through the examinee's body. It is a fairly common technique and can be used in all age categories (Larsson et al., 1984). It belongs to the fast methods, it involves low radiation and does not require special preparation and technical training. By passing dual-energy ionizing radiation through the body, or some of its segments, the attenuation of the signal is recorded, which is a consequence of different tissue thickness, density and chemical composition through which the rays pass. The DXA method can be used to determine FM, lean body mass, and bone mineral density (Mazess et al., 1990). It is primarily used to determine the density of minerals in bones and is considered the gold standard (Garg & Kharb, 2013). It can be used for the analysis of certain parts of the body, as well as for the analysis of the composition of the whole body. In visceral adipose tissue analysis, compared to magnetic resonance imaging, DXA overestimates visceral adipose tissue in obese individuals (Neeland et al., 2016; Lemos & Gallagher, 2017). In the image obtained by DXA scanning, subcutaneous adipose tissue cannot be separated from visceral, but in that case an anatomical model predicting the thickness of subcutaneous adipose tissue must be used. DXA provides a two-dimensional image and it is not possible to directly determine the volume of certain parts, so the use of anatomical models is required (Borga et al., 2018).

2.5 Computed tomography (CT) and magnetic resonance imaging (MRI)

CT and MRI are methods that, by emitting different types and frequencies of radiation, provide high-resolution images in three dimensions. These methods are considered the gold standard, when it comes to body analysis at the tissue level. They have the ability to show even the smallest changes that occur in the body (Prado & Heymsfield, 2014). They can be conducted partially, or on the whole body. In CT, X-rays are transmitted from different angles, in order to obtain an image based on the attenuation of rays, which occurs due to different tissue densities (Addison et al., 2014), so an image of adipose tissue, soft tissue, thickness and volume is obtained. MRI uses the magnetic properties of the body, i.e. the density of hydrogen atoms (they act as a magnet under pulsed radio frequency waves) (Berger, 2002), in order to obtain an image and determine the thickness and volume of adipose and muscle tissue (Silver et al., 2010). Based on this, MRI is a safer method than CT. Due to the high radiation in CT, the practice is to take images of a certain region of the body, not to scan the entire body. Recently, spectroscopic magnetic resonance imaging has played a significant role in the identification of muscle and liver fat, which provides insight into metabolic risks (Hwang & Choi, 2015).

The advantages of these two methods as the most accurate, when it comes to tissue level analysis, are unequivocal, but they are expensive, require trained personnel, they are not easily available and the major disadvantage is the exposure to ionizing radiation in CT. MRI can be a problem for people who have metal implants. On the other hand, MRI can be used in children, because there is no ionizing radiation.

2.6 Ultrasound (US)

A less known technique, in addition to the already mentioned ones in the analysis of body composition, is ultrasound. Most people are familiar with the US as a diagnostic device when it comes to certain health problems or pregnancy, but it is less known that the US is used to measure fat and skeletal muscle thickness (Smith-Ryan et al., 2014). This device creates an ultrasound image, as a result of sound waves that pass through the skin and partially reflect back from tissues, fat and bones in the form of echoes, depending on changes in acoustic resistance in tissue (Wagner, 2013). There are two modes of ultrasound technology, A (amplitude mode) and B (brightness mode). B-mode uses a frequency of 1–10 MHz, while A-mode uses a frequency of 2.5 MHz and these are newer commercial devices (Smith-Ryan et al., 2014). In the obtained ultrasound image, the thickness of the tissue is measured with an electronic calliper. This technique is accessible, non-invasive, fast and there is no radiation, but there is not enough data on the accuracy, when compared to the reference method, so further research is needed. Interest in the analysis of body composition by the US method has grown after the development of portable devices, such as the Body-Metrix ultrasound system, primarily made for use in the fitness industry.

2.7 Bioelectric Impedance Analysis (BIA)

BIA is one of the most effective, fastest, non-invasive, relatively low-cost and valid methods of body composition analysis (Yu et al., 2010). There are a large number of devices on the market, but not all of them are valid. Their validity depends on the quality of the devices, and they are usually expensive, but still significantly more affordable than the previously mentioned methods. This method has also been shown to be a good choice for use in children (Meredith-Jones et al., 2015). There are multi-frequency devices (1, 5, 50, 100, 200, 500, 1000 kHz) and single frequency devices (400 µA and 50 kHz) on the market today (Kyle, Bosaeus, De Lorenzo, Deurenberg, Elia, Gómez, et al., 2004). The difference in measurements between these two types of devices is in the greater ability of multi-frequency devices to perform different measurements, in different positions, to give results for individual body parts (Smith & Madden, 2016). BIA is a method that is based on the body's resistance when passing a very low electric current and a certain frequency through the human body. Electrical conductivity is affected by the water and electrolyte content of the conductors. Higher resistance is provided by tissues with lower water content, and since FFM contains almost all water and electrolytes, its conductivity is far higher than FM. Modern BIA devices provide information on the total amount of water (TBW), intracellular water, extracellular water, the amount of minerals, muscles, fats (total and visceral), as well as data on segmental analysis (all four extremities separately and the trunk), bioelectrical impedance vector analysis (BIVA) and the phase angle (PA) of the whole body. The phase angle of the body has proven to be a very important variable and in healthy individuals it is usually in the range between 5º and 7º. It is considered to show the ratio of intra and extracellular water, that is, of cell health, where higher values indicate better cell function. The phase angle is associated with physical activity, gender (men have higher values due to higher muscle mass), nutrition, inflammation, infection (lower phase angle values) (Norman et al., 2012). Phase angle values can be predictive for many clinical outcomes and mortality in people with chronic diseases (Iqbal, 2013). More detailed data on tissue hydration, body fluid variation, cell mass, as well as longitudinal changes are obtained via BIVA. BIVA provides a qualitative measure of soft tissue, regardless of body size (Norman et al., 2012). Within the 75th percentile are healthy individuals, while values above the 95th are considered abnormal (Walter-Kroker et al., 2011). BIVA helps to interpret changes in body weight and body composition (Nicoletti et al., 2014). The reliability of the BIA method depends on the quality of the device, a well-trained personnel conducting the analysis, accurately measured height and weight, limb position (limb angle of 30º-45º in relation to the body), that the examinee did not eat a few hours before analysis, that the bladder is empty, that the examinee did not exercise, phase of the menstrual cycle, pregnancy, metal or silicone implants, pacemaker (Yamaguchi et al., 2012; Kyle, Bosaeus, De Lorenzo, Deurenberg, Elia, Manuel Gómez, et al., 2004), indoor temperature (24º- 34ºC does not affect measurement results) (Caton et al., 1988; Garby et al., 1990). When there are larger deviations in hydration, the results of FM and FFM should be interpreted carefully, because they can be overestimated or underestimated (Mialich et al., 2014).

CONCLUSION

This mini-review paper highlights the importance of body composition analysis and the most commonly used models and methods. It can be said that so far there is no universal method that can be used to obtain all valid data, but it is necessary to use a combination of methods (multi-component model), in order to obtain valid results. The most accessible and widely used methods are anthropometry and BIA. Both methods are affordable, non-invasive, safe for all age categories, comfortable for the examinee, relatively fast and can be used on-field (some BIA models). In addition to the above, anthropometry is also a cheap method. The other mentioned methods require exclusively laboratory conditions, they are expensive and less comfortable for the examinee, and some of them involve a lot of radiation and they are most frequently used for research purposes. All the mentioned methods require a well-trained personnel to conduct them, with the necessary adherence to the protocol.

As anthropometry is a very accessible and widely used technique, it is necessary to develop techniques in the future that would make it even more precise and reliable and enable segmental analysis of body composition. On the other hand, BIA is an increasingly popular method, it has a wider application and is in constant development. To obtain reliable results, it is recommended to use scientifically validated devices, improve the standardization of measurement protocols and define clinically acceptable limitations in terms of accuracy.

The future concept of body composition analysis should aim to look at the data as a whole, rather than in isolation, to explain the relationship between individual body components, organs and tissues, their metabolic and functional properties, and health risks.

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