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Vücut Kompozisyonu Belirlenmesinde Altın Standart Densitometri Teknikleri Kullanımı

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Özet

Vücut kompozisyonu (VK) değerlendirmesi, sağlık durumu, enerji dengesi ve antrenman–beslenme stratejileri hakkında bilgi sağlayarak klinik uygulamalara rehberlik ve spor performansı takibi temelini oluşturur. Bu derleme, VK'nin antropometri ve iki bileşenli (2B) modelden günümüzde kullanılan çok bileşenli (3B/4B) yaklaşımlara evrimini özetlemekte ve densitometri temelli referans teknikler ile araştırma ve uygulamada yaygın kullanılan ölçüm yaklaşımları değerlendirmektedir. Amaç, model varsayımlarını açıklığa kavuşturmak, yöntemlere özgü hata kaynaklarını özetlemek ve doğruluk ile karşılaştırılabilirliği artıracak standardizasyon gereksinimlerini ortaya koymaktır. Hidrostatik Tartı (HT) ve Hava Yer Değiştirme Pletismografisi (HYDP), laboratuvar ortamında vücut hacmi belirlemede standart yöntemler olarak incelenmiş, akciğer hacmi düzeltmelerinin (HT'de rezidüel volüm, HYDP'de Torasik Gaz Volüm) kritik etkisi vurgulanmıştır. DXA'nın kemik mineral içeriği, yağ ve yağsız kütle tahminlerindeki rolü ele alınmış; cihaz/yazılım farklılıkları, hidrasyona bağlı değişkenlik ve sporcu ölçümleri üzerindeki etkileri tartışılmıştır. Sporcu popülasyonlarından elde edilen kanıtlar, yağ kütlesi (YK), yağ harici kütle (YHK) ve vücut yağ yüzdesi (%VY) ölçümlerinde yöntem bağımlı farklılıkları göstermiş ve çok bileşenli modellerin sonuçların güvenirliğini artırdığı vurgulanmıştır.Sonuç olarak, densitometri (HT, HYDP) ve DXA temel referans yöntemler olmaya devam etmektedir; ancak bu yöntemlerin altında yatan varsayımlar ve metodolojik tercihler, elde edilen tahminleri önemli ölçüde etkilemekte ve farklı popülasyonlarda doğruluğun sağlanabilmesi için süreğen standartlaştırma ve iyileştirme gerekliliğini ortaya koymaktadır. Bu tekniklerin dört bileşenli modeller, uyumlu test protokolleri, popülasyona özgü tahmin denklemleri ve boylamsal takip eşikleri (en küçük saptanabilir değişim, klinik olarak anlamlı en küçük fark) ile bütünleştirilmesi, ölçüm hatasını en aza indirecek, çalışmalar arası karşılaştırılabilirliği artıracak ve daha sağlam klinik ve spora özgü karar verme süreçlerini destekleyecektir.

Anahtar Kelimeler: *Vücut Kompozisyonu, Dansitometri, Hidrtostatik Tartım, Hava Yer Değiştirme Pletismografisi (HYDP), Dört Bileşenli(4B) Model*

Use of Gold Standard Densitometry Techniques in the Determination of Body Composition

Abstract

Body composition (BC) assessment underpins clinical practices guidance and sport performance follow up by informing health status, energy balance, and training–nutrition strategies. This review synthesizes the evolution of BC—from early anthropometry and the two-component (2C) model to contemporary multi-component (3C/4C) approaches—and evaluates densitometry-based reference techniques alongside widely used measurement approaches in research and practice. The aim is to clarify model assumptions, summarize method-specific sources of error, and outline standardization needs to improve accuracy and comparability. Hydrostatic weighing (HW) and air displacement plethysmography (ADP) are examined as laboratory standards for determining body volume, emphasizing the critical influence of lung-volume corrections (residual volume in HW, thoracic gas volume in ADP). DXA's role in estimating bone mineral, fat, and lean mass is reviewed, highlighting device/software variability, hydration effects, and implications for athlete assessment. Evidence obtained from athlete populations has demonstrated method-dependent differences in fat mass (FM), fat-free mass (FFM), and body fat percentage (%BF) measurements, and has emphasized that multi-component models increase the reliability of the results. In conclusion, densitometry (HW, ADP) and DXA remain primary reference methods, but their underlying assumptions and methodological choices significantly influence the resulting estimates, underscoring the need for ongoing standardization and refinement to ensure accuracy across diverse populations. Integrating these techniques with four-component models, harmonized testing protocols, population-specific prediction equations, and longitudinal thresholds (smallest detectable change, minimal clinically important difference) will minimize measurement error, enhance cross-study comparability, and support more robust clinical and sport-specific decision-making.

Key Words: *Body Composition, Densitometry, Hydrostatic Weighing, Air Displacement Plethysmography (ADP), Four Compartment (4C) Model*

Introduction

Accurate and reliable measurement and evaluation of body composition (BC) provide critical information regarding overall health status, physical fitness, athletic performance, and an individual's energy balance. Proper determination of body weight in relation to ideal body components, along with the identification of excessively low or high BF%, facilitates the achievement of an optimal BC. In this context, BC assessments serve not only as a tool for individual health monitoring but also as a crucial basis for designing exercise programs and developing nutritional strategies. Particularly in relation to multidimensional goals such as enhancing athletic performance, improving metabolic health indicators, and reducing the risk of chronic diseases, the accurate evaluation of BC through appropriate methods is of paramount importance (Nana, Slater, Stewart, & Burke, 2015). Furthermore, body weight remains a critical factor in exercise sciences, as it determines athlete classification and competition categories (e.g., wrestling, judo, boxing). In endurance sports, such as long-distance running, maintaining a low BF% is particularly relevant given the load carried by the body. Accordingly, body structure and composition are well established as key determinants of athletic performance (Wilmore, 1979), and the assessment of body composition is therefore recognized as a essential tool in both sports sciences and clinical health applications (Ackland et al., 2012; Wells & Fewtrell, 2006).

The Importance and Historical Development of Body Composition

The study of BC can be traced back to ancient times, with the earliest conceptual approaches appearing in the 4th century BC during the Ancient Greek period. Nevertheless, the foundations of systematic and scientific investigations in the modern sense were laid in the mid-19th century and further advanced in the early 20th century, evolving into an interdisciplinary field (Brožek & Henschel, 1961). By the early 1900s, subcutaneous fat was estimated through skinfold thickness measurements, with subsequent studies reporting strong correlations among different body sites. In the 1960s and 1970s, anthropometric equations were introduced to estimate total body density and BF% (Durnin & Womersley, 1974; Jackson & Pollock, 1978; Jackson, Pollock, & Ward, 1980), though these methods were limited in both accuracy and practicality (Behnke, Feen, & Welham, 1942; Brožek & Henschel, 1961). A major milestone was achieved in the early 1940s with Behnke's development of the two-component (2C) model, together with HW, which provided the first systematic distinction between FM and FFM (Behnke et al., 1942; Brožek & Henschel, 1961; Buskirk, 1959). This model laid the foundation for subsequent conceptual and methodological advances in BC research.

Within this historical advancement, various techniques were developed to estimate BC through indirect methods. In particular, the assessment of BF% using skinfold thickness measurements became widely adopted in both field applications and scientific research beginning in the early 20th century. The equations developed to estimate body density and BF% through anthropometric measurements (Jackson & Pollock, 1978; Jackson et al., 1980; Jackson, Pollock, Graves, & Mahar, 1988) facilitated the widespread use of this method, owing to its ease of application and cost-effectiveness. Moreover, population-specific equations developed for children, adolescents, adults, and athletic population have provided scientific validity for age- and sex-specific assessments (Jackson & Pollock, 1978; Jackson et al., 1980; Jackson et al., 1988; Heyward & Wagner, 2004; Küçükkubaş, 2007; Küçükkubaş, Aytar, Açıkada, & Hazır, 2020; Lohman, 1981; Norton & Olds, 1996; Slaughter et al., 1988; Sloan, 1967; Withers, Craig, Bourdon, & Norton, 1987). Building on these advances, contemporary prediction equations now incorporate additional anthropometric variables such as girth and breadth measurements (Heyward & Wagner, 2004). Collectively, these developments provided the conceptual and methodological cornerstone for modern indirect and doubly indirect approaches to BC assessment.

Body Composition Assessment Methods and Concepts

The only method that allows direct and precise measurement of BF% is cadaver dissection analysis (Behnke, 1959a). However, due to ethical and practical constraints, this method has been applied in only a limited number of studies to date (Heyward & Wagner, 2004). Consequently, all other techniques used for the assessment of BC fall into the categories of indirect or doubly indirect methods (Wang, Pierson, & Heymsfield, 1992). The measurement and evaluation of BC represent a key factor widely implemented in both research and clinical applications. The use of fundamental concepts and terminology often leads to errors in evaluation, comparison, and interpretation. To clarify key concepts, standardize terminology, and provide guidance on the application and interpretation of BC assessment, the referenced article focused on methodological standards, summarized BC levels and models, and presented definitions using standardized terms (Prado et al., 2025). At the molecular level, the most commonly used model is the 2C model, which is based on FM and FFM. This model is applied in methods such as ADP and HW (hydrodensitometry/underwater weighing) (Prado et al., 2025).

A wide range of methods are used in the assessment of BC, each differing in their scientific foundations, levels of accuracy, and conditions of applicability (Brožek & Henschel, 1961). Among these methods are Hydrostatic Weighing (HW), Air Displacement Plethysmography (ADP), Isotope Dilution, Dual-Energy X-Ray Absorptiometry (DXA), Potassium-40 Counting, Neutron Activation Analysis, Bioelectrical Impedance Analysis (BIA), anthropometric measurements, Near-Infrared Interactance (NIR), and Magnetic Resonance Imaging (MRI) (Carey, 2000; Fields & Goran, 2000a; Heyward & Gibson, 2014; Nielsen et al., 1993; Roemmich, Clark, Weltman, & Rogol, 1997; Salmi, 2003; Wagner & Heyward, 1999). In the literature, some of these methods are regarded as reference measurement techniques, and accordingly, various prediction equations have been developed (Slaughter et al., 1984). At the same time, there are also comparative studies examining the agreement or discrepancies among different measurement techniques (Küçükkubaş et al., 2020; Thomas, Crofford, Scudder, Oletti, Deb, & Heymsfield, 2025). In such studies, significant differences have been identified between certain techniques (Boye, 2002; Dalsky et al., 1990; Sopher et al., 2004; Küçükkubaş et al., 2020; Wang, Heymsfield, Aulet, Thornton, & Pierson, 1989), while other research has reported no statistically significant differences among methods (Heymsfield, 1989; Mazess, Peppler, & Gibson, 1984; Thomas et al., 2025). Notably, neutron activation analysis, which is employed to enhance the accuracy of multicomponent models, although highly sensitive, is associated with considerably high costs. Among the most technically complex and challenging methods are Oxygen^{18} Dilution, Neutron Activation, and MRI. On the other hand, ADP, HW, and neutron activation analysis are highlighted among the techniques reported to provide the most accurate assessment of BC. Within the framework of BC assessment, a variety of methods and conceptual models have been developed to improve the accuracy of estimating FM and FFM across populations. Among these, the 2C model has historically provided the fundamental framework by partitioning body mass into fat and fat-free compartments, forming the basis for the development of more advanced, multi-component approaches.

Two-Component Model (2C): Theoretical Basis, Assumptions, and Limitations of the Reference Body Concept

In BC analysis, models are generally examined as chemical models and whole-body models. One of the classical and most frequently applied approaches is the 2C model, which divides the body into two main compartments: FM and FFM. In this approach, FM encompasses all extractable body fats, while FFM consists of water, protein, and mineral components (Wang et al., 1992; Siri, 1961; Siri, 1961; Wells & Fewtrell, 2006). The assumptions of this model were derived from cadaver studies, which provided the first empirical estimates of body compartment densities. Cadaver dissections of three white males aged 25, 35, and 46 led to the establishment of the so-called “reference body,” from which the densities of FM (0.901 g/cm^3) and FFM (1.100 g/cm^3) were determined (Behnke, 1959a,1959b; Luft & Lim, 1959). On this basis, the 2Cmodel assumes that these density values apply uniformly to all individuals and that, within FFM, both the component densities and their relative proportions (e.g., water, protein, mineral) are invariant across individuals. However, longitudinal evidence indicates that not only the hydration of fat-free tissues but also their composition (e.g., age-related shifts in bone mineral and protein content) varies during growth and maturation, challenging these constancy assumptions—particularly in pediatric populations (Wells & Fewtrell, 2006). These limitations have underscored the need for more advanced methodologies in BC research. In recent years, technological advancements have greatly expanded the possibilities for measuring human BC. Since the components of BC vary across populations depending on factors such as age, sex, genetic background, ethnicity, and environmental influences, these variations directly affect the accuracy of measurement techniques and the validity of prediction equations. Such population-specific variability may particularly constrain the standardization and applicability of indirect methods such as anthropometry and bioelectrical impedance analysis (BIA). Therefore, to ensure high validity in BC analyses, prediction equations derived from reference methods and tailored to the characteristics of the target population are increasingly employed (Wells & Fewtrell, 2006; Marin-Jimenez et al., 2022).

Despite its limitations, the 2C model continues to be widely used because of its simplicity, cost-effectiveness, and applicability in both clinical and field settings. Nevertheless, recognition of its inherent assumptions and methodological constraints has driven the development of more advanced multi-component approaches—most notably the three-component (3C) and four-component (4C) models—which provide greater accuracy by explicitly accounting for variability in body water and bone mineral content."

Densitometry-Based Reference Methods: HW and ADP

A comprehensive evaluation of BC requires consideration of densitometry-based techniques, which, despite methodological limitations, have historically been regarded as the gold standard in body composition assessment and continue to serve as reference methods and benchmarks for validating other techniques. Among these, HW and ADP represent the principal densitometric approaches and are used to validate other indirect methods. Nevertheless, recognition of the inherent assumptions and constraints of these methods has driven the development of multi-component models. The 4C model, in particular, partitions the body into fat, water, protein, and mineral, thereby reducing error. However, due to practical challenges in its application, there remains a demand for simpler alternatives. In this context, the 2C model continues to warrant discussion, as it provides a balance between feasibility and acceptable levels of precision and accuracy.

Given the practical advantages of the ADP over HW—such as avoiding submersion in water, eliminating the need for maximal underwater exhalation, reducing measurement time, and requiring no tank maintenance—it is not surprising that ADP has largely replaced HT as the laboratory method for determining body volume (Fields, Hunter, & Goran, 2000b; McCrory, Gomez, Bernauer, & Mole, 1995). Indeed, a search of the Scopus database combining the term "body composition" with "air displacement plethysmography" yields nearly five times as many publications over the past 20 years compared with searches using "hydrostatic weighing" or "hydrodensitometry" (Scopus, 2024). This trend highlights ADP's current status as one of the preferred reference methods for body composition research.

Among these densitometric techniques, HW has historically been regarded as the classical gold standard. A closer examination of HW—and the critical role of residual lung volume (RV)—provides a foundation for the subsequent discussion of the other densitometric method, ADP, and its specific consideration of thoracic gas volume (TGV).

"Hydrodensitometry" – "Hydrostatic Weighing" – "Underwater Weighing"

In the literature, Hydrostatic Weighing (Underwater Weighing or Hydrodensitometry) has long been regarded as the gold standard method for determining body density (D_b) with high precision (Heyward & Gibson, 2014; Heyward & Wagner, 2004; Ackland et al., 2012). Nevertheless, advanced technological methods such as DXA, ADP (or BOD POD), and 4C models are also frequently referenced for their accuracy and applicability (Fields, Goran, & McCrory, 2002; Wang et al., 1992). However, HW continues to serve as a primary reference in comparative studies, owing to its physical principles, low margin of error, and ability to directly measure D_b (Ackland et al., 2012).

D_b is determined by relating total body mass to body volume. The HW method is frequently used to determine body volume, and this technique is based on Archimedes' Principle (Heyward

& Gibson, 2014). Although it is a 2C model, D_b measurement through HW is considered the "gold standard" for BC assessment (Ellis, Pratt, Puyau, Shypailo, & Butte, 2001a; Ellis & Shypailo, 2001b; Salmi, 2003). Based on the 2C model, several different measurement techniques have been developed for BC assessment. Using the densitometry method, D_b is calculated according to the 2C model, and BF% is then estimated. Although the HW method is regarded as the gold standard, it is costly and requires effective communication skills between the measurer and the subject (Kushner, Schoeller, Fjeld, & Danford, 1992) and with multiple sources of error and limitations, and it is often considered impractical by many investigators.

In HW, subjects must be submerged in water to measure total D_b. During immersion, the subject is required to perform complete expiration, a maneuver that demands strong cooperation between the subject and the examiner. In general, this procedure is repeated until three consecutive measurements are obtained within approximately 100 g of each other. The average of these three measurements is then used to calculate body volume and D_b (Bonge & Donnelly, 1989). The comparison of the subject's HW and normal body weight, along with the volume of water displaced, is used to calculate overall D_b. This result is then corrected for residual lung volume. The residual volume is most commonly measured using the oxygen dilution technique with a closed-circuit spirometry system (Goldman & Buskirk, 1959; Salmi, 2003).

Although HW method can measure body weight and volume with high accuracy, certain assumptions regarding tissue density must be taken into account when estimating BF%. In particular, individual variations in body fluid content, as well as protein and mineral levels, may lead to errors in estimation. The total error rate in predicted BF% is generally around 3–4% of an individual's body weight (Heyward & Gibson, 2014). Therefore, considering that variations in FFM and body fluids may influence measurement results, it is recommended that densitometric methods not be used as reference techniques in heterogeneous groups (Salmi, 2003). A similar situation has been documented in methods used to assess children's growth status, where uncertainties exist regarding which region, technique, or instrument would provide the most meaningful data (Lohman, Roche, & Martorell, 1988). In children, equations have been developed for both the 4C model based on HW (Roemmich et al., 1997; Sopher et al., 2004; Wells et al., 1999) and for reference methods applied to children and adolescents (Ellis, Shypailo, Abrams, & Wong, 2000). To minimize the assumptions and error rates associated with these techniques, new regression equations have been developed by directly measuring the body's four separate components and combining them with densitometry.

D_b is estimated through HW using specific prediction equations (Brozek, Grande, Anderson & Keys, 1963; Siri, 1961). The obtained D_b value is then converted into BF% (%BF) through additional equations, the reliability of which is expected to be at least as high as that of the original D_b equations (Wagner & Heyward, 1999). Two-component models typically estimate FM and FFM. However, the density of FFM varies according to age, sex, body fatness, and physical activity level (Baumgartner, Heymsfield, Litchman, Wang & Pierson, 1991; Wang et al., 1989). Therefore, population-specific density equations should be applied (Table 1). Numerous equations based on BD have been developed for different populations (Heyward & Wagner, 2004). In addition to equations designed for adults (Jackson & Pollock, 1978; Jackson et al., 1980; Sloan, 1967), specific equations have also been developed for children (Slaughter et al., 1988; Boileau, Lohman & Slaughter, 1985; Boileau, Wilmore, Lohman, Slaughter & Riner, 1981; Lohman, 1981; Lohman, 1986).

Any discrepancy between the assumed and the actual values of FFM density can lead to substantial differences in the estimation of BF%. For example, if an individual's overall D_b is 1.07 g/ml, the predicted BF% using Siri's equation is 12.6%. However, if the actual density of FFM is 1.12 g/ml instead of 1.10 g/ml, the correct BF% would be 19.1%. Conversely, if the FFM density is 1.08 g/ml, the true BF% would be 4.7%. When compared to adults, children typically have a higher water content and lower bone mineral density within their FFM. As a result, FFM undergoes greater variation during growth. The range of estimation in boys, for instance, can vary from 1.063 g/ml at birth to 1.100 g/ml (Lohman, Boileau & Slaughter, 1984). Therefore, two different body fat equations have been developed for these populations (Table 1). Ultimately, there remains a need for equations that are derived from D_b measurements and that are simple to apply while minimizing error.

Building on the need for population-specific prediction equations, various debates have centered on the selection, measurement, and evaluation of the reference method used to derive such equations. Lohman (1981) argued that prediction equations should primarily be developed for D_b. Lohman (1981) argued that prediction equations should primarily be developed for D_b. He further emphasized that equations for FFM, FM, and fat content should also be established, and that population-specific variations in BC should be taken into account when deriving such equations. Including such information in studies would provide a significant advantage. This, in turn, would allow the calculation of BC in individuals who do not fit the reference BC (e.g., women, children, elderly men, mesomorphic athletes, and obese populations) by applying equations other than those of Siri (1961) or Brozek (1963) to estimate FFM, FM, or fat content from D_b values. These considerations underscore the importance of empirical studies that evaluate the applicability and accuracy of existing prediction equations across diverse populations—such as women, children, elderly men, mesomorphic athletes, and obese groups and against different reference methods.

In a recent study, BF% was measured in premenopausal Hispanic women (n = 78) using HW (with Siri, Brozek, and Lohman equations) and DXA, another major reference method. The researchers performed linear regression and Bland–Altman analyses to compare the two methods and found no significant differences between the mean values. Although this study focused on evaluating the accuracy of existing 2C equations rather than developing new regression models, it serves as a representative example of how reference methods are compared in specific populations. In this context, the selection of reference techniques remains a critical issue in BC research. This becomes particularly evident when considering the 2C model, which assumes that the BC of the individual being measured differs from the reference body model only in terms of fat content. Within this framework, FFM in the reference body is presumed to consist of approximately 73.8% water, 19.4% protein, and 6.8% minerals (Heyward & Wagner, 2004; Wang et al., 1992). Put differently, the composition of all non-fat tissues in an individual is assumed to closely reflect these proportions. Guided by these assumptions, BF% is calculated through indirect measurement methods based on D_b. However, because these assumptions are sensitive to individual differences such as age, sex, ethnicity, and physiological development, the model may compromise accuracy across diverse populations. In this regard, one of the major methodological considerations in HW is the accurate measurement of residual lung volume, which can substantially affect D_b calculations (Morrow, Jackson, Bradley, & Hartung, 1986; Wagner & Heyward, 1999; Wilmore, Vodak, Parr, Girandola, & Billing, 1988b).

Residual Volume Considerations in Hydrostatic

HW, although one of the most widely applied densitometric methods, poses significant challenges for field application due to its demanding and costly equipment requirements. In particular, potential errors in the estimation or direct measurement of residual lung volume (RV) may compromise the accuracy of results. Since D_b is defined as the ratio of body mass (BM) to body volume, and D_b is used to predict BF%, accurate assessment of RV is essential during HW (Wagner & Heyward, 1999).

HW is based on Archimedes' principle, which states that the loss of weight in water is equal to the volume of displaced fluid. Accordingly, body volume can be determined by recording water displacement with a volumeter or burette (Wagner & Heyward, 1999). Alternatively, density can be calculated directly from mass and volume (Behnke & Willmore, 1974). Therefore, RV must always be assessed to ensure validity (Morrow et al., 1986; Wagner & Heyward, 1999). When comparing measured RV values with estimated values, prediction errors of approximately 2.85% in women and 3.7% in men have been reported (Morrow et al., 1986). RV is typically measured using closed-circuit helium dilution or oxygen dilution techniques (Wilmore et al., 1988a).

Accordingly, density is defined as mass/volume. If body volume can be measured, D_b can be calculated using the following equation:

$$Vb = Wa / (((Wa - Ww) / \rho w) - RV - 0.1)$$

where Vb = Body Volume; Wa = Body weight in air; Ww = Body weight in water; ρw = Water density; RV = Residual lung volume; 0.1 = Gastrointestinal gas volume (L) (Behnke & Willmore, 1974). In this equation, the key factors are RV, gastrointestinal gas, and the density of water. Once D_b has been determined, BF% can then be estimated using key equations derived from D_b (Table 1).

HW and RV application procedures require highly detailed preparation and execution (Goldman & Buskirk, 1959; Heyward & Gibson, 2014; Heyward & Wagner, 2004; Maud & Foster, 1995; Norgan, 1991). The establishment of the necessary equipment for HW measurements in the laboratory, as well as issues such as hygiene, are both costly and demand a high level of expertise, typically requiring teamwork by specialized personnel. Studies have reported significant inter-individual differences in RV, with values ranging from 0.996 to 1.965 liters (Marks & Katch, 1986). In a preliminary doctoral study, RV measured using the head-out-of-water technique was found to be higher than RV measured underwater with the head submerged. While repeated in-water (head-submerged) trials showed no statistically significant differences across repetitions according to ANOVA, direct comparisons between head-out and in-water measurements revealed significant discrepancies. Based on these findings, the investigators concluded that RV should be measured simultaneously during underwater weighing with the head submerged (Küçükkubaş, 2007).

These methodological challenges highlight the critical impact of RV assessment on the accuracy of BC outcomes, as even minor errors in its estimation can substantially alter the calculation of BF%. For example, even small errors in RV measurement can lead to considerable inaccuracies in body fat estimation; a miscalculation of only 100 ml in RV may shift BF% by nearly 1%. In this context, Morrow et al. (1986) compared the accuracy of D_b determined from measured versus predicted RV values in 46 adults and 134 athletes. For RV measurement, either the oxygen dilution technique or the nitrogen washout technique was employed. In predicting RV, variables such as height, age, and vital capacity were used. However, when RV was estimated rather than directly measured, HW resulted in substantial errors in BF%, corresponding to 3.7% in men and 2.85% in women.

Given these limitations of HW, alternative densitometric methods have been developed, among which ADP has emerged as a widely adopted approach.

Air Displacement Plethysmography (ADP) and the BOD POD® System

ADP system was first introduced in the mid-1990s, with initial publications outlining both the theoretical foundations of the method (Dempster & Aitkens, 1995) and its validation against HW (McCrory et al., 1995). Compared with HW, ADP offers several practical advantages, including shorter testing time, no water immersion, elimination of maximal exhalation underwater, and reduced equipment maintenance. Owing to these advantages, ADP has progressively replaced HW as a laboratory standard for determining body volume. Indeed, bibliometric analyses indicate that over the past two decades, the number of studies employing ADP has been several times greater than those utilizing HW.

It is a valid and reliable method for measuring an individual's body volume. In this technique, the subject sits inside a closed chamber (commonly the BOD POD), and changes in air pressure within the chamber are measured to calculate body volume based on Boyle's law. The system consists of two chambers—a test chamber containing the subject and a reference chamber—between which small pressure oscillations are applied. The resulting pressure–volume relationships are analyzed to determine body volume. To minimize air displacement errors caused by clothing and hair, participants are required to wear tight-fitting swimsuits or spandex garments and a swim cap. Once body volume has been obtained, D_b is calculated as body mass divided by body volume, and BF% is subsequently estimated using standard equations. Compared with HW, ADP eliminates the need for water immersion and maximal exhalation underwater, thereby reducing participant burden and measurement error while maintaining comparable accuracy (Dempster & Aitkens, 1995; McCrory et al., 1995; Fields et al., 2000b).

ADP is particularly advantageous in populations such as children, older adults, and clinical cohorts due to its non-invasive nature, rapid application, and absence of radiation exposure. Initially developed as an alternative to HW, the method has been validated in several studies as providing comparable accuracy (Fields et al., 2000b). Nonetheless, a methodological limitation of ADP lies in its sensitivity to clothing and hair, as garments other than tight-fitting swimsuits or uncovered hair may cause air displacement errors, thereby compromising the accuracy of D_b estimates (Fields et al., 2000b). Accordingly, standardized testing protocols mandate the use of form-fitting garments and a swim cap to ensure validity in measurement outcomes. Beyond issues related to clothing and chamber conditions, the consideration of thoracic gas volume (TGV)—rather than residual volume as in HW—represents a critical methodological factor for ensuring valid ADP outcomes.

Thoracic Gas Volume Consideration in Air Displacement Plethysmography (ADP)

Thoracic Gas Volume (TGV), a critical determinant in D_b calculations, constitutes one of the major potential sources of error in BC assessment and may be obtained either through direct measurement or prediction equations. In BOD POD –based validation studies, RV assessment has therefore been performed using both approaches. Unlike HW, which relies on maximal exhalation to residual volume, the ADP generally assesses lung volume based on the average level during normal tidal breathing. Accurate assessment of TGV in ADP systems, is critical for valid estimation of D_b and BF%. Accordingly, research using the ADP has frequently examined the agreement between measured and predicted TGV values to assess the accuracy of this methodological choice; recent investigations continue to highlight systematic biases that vary by population and prediction formula. Miller (2020) reported no significant differences between measured and predicted TGV values in college-aged students, supporting the use of predicted equations when direct measurement is not feasible. However, previous studies have highlighted population-specific discrepancies, particularly in athletes and children, suggesting that reliance solely on prediction equations may compromise accuracy in certain groups (McCrory et al., 1995; Minderico, et al., 2006; Collins et al., 1999).

Ducharme, Gibson, and Mermier (2021) reported that the accuracy of predicted TGV decreases as measured TGV increases, leading to systematic underestimation in individuals with larger lung volumes. This bias was particularly evident in men with measured TGV ≥ 4.5 L and women with measured TGV ≥ 3.5 L, which resulted in underestimation of both TGV and %BF. Their findings highlight sex as an important covariate influencing prediction accuracy, independent of height. Accordingly, the authors recommend that TGV should be directly measured whenever possible in adults aged 18–30 years, as reliance on predicted values may compromise validity in young populations, especially in men. More recently, a study directly compared the three most commonly used TGV prediction formulas—Crapo, Fields, and Ducharme —in young adult athletes (Wagner et al., 2025). The findings indicated that the Ducharme equation was the only formula that produced mean values not significantly different from measured TGV, and it also yielded fewer large errors (≥ 2% BF) compared to Crapo and Fields. Nevertheless, all three formulas demonstrated systematic bias, with overestimation occurring in individuals with small TGV and underestimation in those with large TGV.

Although Ducharme attenuated the magnitude of this error relative to Crapo, the authors emphasized that direct measurement remains preferable whenever accuracy is critical (Wagner et al., 2025). Taken together, these studies indicate that prediction equations for TGV, while useful in practice, are limited by population- and sex-specific biases, underscoring the methodological importance of direct measurement. Looking ahead, future research should prioritize refining sex- and population-specific prediction models and validating them across diverse groups, while continuing to treat direct TGV measurement as the reference standard when precision in BC assessment is required.

Why the Four-Component (4C) Model?
The Roles of Densitometry and DXA

In recent years, methods aimed at achieving more precise assessments of BC have gained increasing importance (Wagner & Heyward, 1999). 4C model was developed to independently evaluate fat, body water, protein, and bone tissue, thereby minimizing the errors that result from assuming a constant composition of FFM. The integration of DXA—used to measure bone mineral content—with techniques for assessing body water is a key methodological advance that enhances the accuracy of the model. This combined approach underscores the value of directly measuring bone mineral and body water to generate more accurate and population-specific estimates of BC.

Various debates exist regarding the selection, measurement, and evaluation of the reference method used for developing prediction equations. Building on this premise, Lohman (1981) emphasized that population-specific differences—such as hydration or bone mineral content—should be integrated into prediction equations to ensure more accurate estimates for diverse groups (e.g., women, children, elderly, athletes, obese populations). This approach would allow the calculation of BC in individuals who do not fit the reference BC (e.g., women, children, elderly men, mesomorphic athletes, and obese populations) by applying equations other than those of Siri (1961) or Brozek (1963). This perspective has become increasingly relevant, as the limitations of 2C models have underscored the need for more advanced approaches, such as the 4C model, which directly measures key components like bone mineral and body water to minimize error.

In this context, DXA has emerged as a widely accepted reference method, offering precise assessment of bone mineral content—one of the key population-specific variables influencing BC estimates—and thus plays a critical role in improving the accuracy of multi-compartment models.

DXA as a Reference Method and Its Role in Bone Tissue Assessment

In BC assessment, although the components in 4C models may appear fixed, their definition and measurement can vary depending on the techniques employed or the purpose of the model. Multi-compartment models, by integrating data from multiple laboratory methods, minimize the errors associated with the assumptions of 2C model (Wagner & Heyward, 1999). In the 4C model, measurements of D_b, water, bone mineral, and FM are combined. Because 2C of FFM—bone mineral and water—are directly measured, this approach represents the most accurate method for determining D_b. When measurement error is minimized, the estimation error for BF% may fall below 2%. Consequently, the 4C model enables the development of the most precise prediction equations for specific populations. For example, in children, body water–density prediction equations have proven highly effective for estimating fat content, allowing more accurate fat assessments and yielding population-specific insights (Lohman, 1986). Furthermore, Sopher et al. (2004) used the 4C model as a criterion standard in children aged 6–18 years and compared it with DXA-derived fat percentage values. Although the results differed slightly, a strong correlation (r = 0.85) was observed, reinforcing DXA's value as a practical clinical tool for BC assessment.

DXA is now widely accepted as a reference method for BC assessment via whole-body scans (Gonera-Furman, Bolanowski, & Jędrzejuk, 2022). Compared to HW, DXA is more frequently utilized because it offers several advantages: measurements are relatively fast, technical error is minimized, and participants are exposed to very low radiation doses. The principle of DXA analysis is based on the differential attenuation of X-rays at two distinct energy levels, which vary according to tissue density and composition, and are combined to produce an attenuation ratio (R value). Whole-body DXA scans with modern densitometers deliver a radiation dose of approximately 4–5 µSv—lower than daily natural background exposure—making the method safe for repeated use (Gonera-Furman et al., 2022). Approximately 40–45% of the pixels in a DXA scan contain bone, meaning that fat and lean mass estimates are derived from the remaining ~60% of body pixels. Importantly, DXA provides whole-body and regional estimates of bone mineral content, FM, and FFM, which enhances its value in both clinical and research settings.

Despite these advantages, several methodological considerations must be acknowledged. DXA assumes that X-ray attenuation coefficients are constant across individuals; however, tissue densities vary by age, sex, ethnicity, and training status. Williams et al. (2006), in a cohort of 215 participants including healthy, obese, and diseased subjects, reported that DXA bias varied systematically with age, sex, body size, and disease state, suggesting that DXA may be unreliable for case–control or longitudinal studies when substantial changes in nutritional status occur between measurements. The coefficients used by DXA software are also influenced by tissue hydration levels, and it is assumed that pixels containing bone have the same fat content as non-bone pixels—an assumption that may not always hold true (Gonera-Furman et al., 2022). Additionally, DXA results may be affected by hydration status, potentially leading to overestimation of FFM and underestimation of FM. Inter-device variability (e.g., pencil-beam vs. fan-beam) and differences in software algorithms can further contribute to discrepancies (Bilsborough, Greenway, Opar, Livingstone, & Coutts, 2014; Njoku, Obasi, Okposhi, Anwara, & Stewart, 2023).

These methodological limitations have significant practical implications in athletic populations, where precise and reliable assessment of FM and FFM is essential for optimizing performance, informing weight-class decisions, and supporting evidence-based training and recovery strategies. This underscores the importance of addressing sport-specific considerations when interpreting DXA-derived body composition data.

Methodological Considerations in Sport-Specific Body Composition Assessment

Sport-specific profiles consistently show meaningful differences in FFM, FM, and BF%. For example, 4C assessments in university club athletes document substantial between-sport heterogeneity in BF% and FFM when a criterion multi-compartment model is used, underscoring that single-method norms may mask sport-driven variation (Wagner et al., 2025). In combat sports, DXA-based reference data and cross-method comparisons indicate typical male BF% centiles around the mid-teens, with method choice (anthropometry vs. BIA vs. DXA) materially shifting absolute estimates (Baranauskas et al., 2023; Dimitrijevic et al., 2022). Across athlete samples, DXA remains the most commonly adopted reference in practice, yet BF% from DXA often reads higher than other techniques, and agreement with field methods is imperfect: BIA tends to overestimate FFM relative to DXA with wide limits of agreement, cautioning against interchangeable use in athletes (Dzator et al., 2023). Although BIA is widely used due to its practicality, prediction equations may not be interchangeable across populations, particularly in children and adolescents, where hydration status and body proportions vary significantly (Deurenberg et al., 1990; Schaeffer et al., 1994; Sun et al., 2003). In a study on youth athletes, anthropometric and BIA-derived estimates of body fat and fat-free mass were significantly different, suggesting that these methods cannot be used interchangeably (Harbili, 2008). Furthermore, hydration status and menstrual cycle phase have been shown to significantly influence impedance values, body fat, FFM, and total body water estimates, limiting the reliability of BIA under non-standardized conditions (Hazır et al., 2003). Seasonal tracking studies likewise show that method selection influences detection of small, position- or phase-related changes in FM/FFM, again favoring standardized protocols and, where feasible, multi-compartment models for inference (Uchiyama et al., 2023). Consensus guidance therefore recommends prioritizing validated laboratory methods (DXA, and ideally multi-compartment models) for decision-making, reserving single-frequency BIA or anthropometry for screening/monitoring only when anchored to sport- and sex-specific equations and calibrated against a reference (Mathisen et al., 2023). In sum, both the magnitude and direction of between-sport differences in FFM, FM, and BF% are method-dependent; using DXA (and preferably 4C) strengthens comparability, reduces bias, and yields more defensible conclusions for sport-specific profiling and longitudinal follow-up. However, as highlighted by Williams et al. (2006), DXA bias is not uniform and varies with age, sex, body size, and disease state, indicating that caution is warranted when using DXA alone for case–control or longitudinal investigations, particularly in populations undergoing major changes in nutritional status.

Despite the dominance of DXA and BIA in recent sport-specific investigations, other densitometric techniques remain relevant for their methodological rigor. Although the BOD POD has been widely validated as a reliable method for estimating body volume and BC, recent studies have shown a clear trend toward the use of DXA or BIA due to their accessibility, ability to provide regional estimates, and suitability for large-scale testing protocols (Nana et al., 2020; Tinsley et al., 2020; Friesen et al., 2022). Nevertheless, the ADP continues to offer distinct methodological strengths, particularly when accurate and rapid assessment is required in athletic populations.

Beyond the widespread adoption of DXA and BIA, HW retains its role as a foundational reference technique in sport-specific BC research, especially in studies requiring precise validation of alternative methods. For example, Larson et al. (2021) examined master swimmers and compared HW with ADP and BIA, confirming HW's accuracy as a criterion method in aquatic athletes where buoyancy plays a key role in performance. Similarly, Jagim et al. (2023) utilized a 3C model—combining HW and total body water—to validate field and laboratory techniques across diverse athletic populations, including strength- and endurance-based sports, underscoring HW's relevance in both performance monitoring and research validation. More recently, Kapuš et al. (2024) conducted a pilot study focusing on BC and buoyancy, again applying HW to assess reliability in aquatic contexts, which is particularly relevant for sports such as swimming and water polo. Collectively, these studies highlight that while the logistical demands of HW limit its frequent use in contemporary practice, it continues to provide a valuable benchmark for validating emerging techniques and ensuring precision in athlete-specific assessments. Its role is especially pertinent in sports where BC directly impacts performance, such as swimming, endurance running, or weight-category sports, thereby reinforcing the methodological importance of maintaining HW as a reference tool despite its practical challenges.

Alongside HW, ADP, commonly implemented using the BOD POD, has also contributed valuable insights in recent years, particularly when integrated with DXA or used for cross-method validation in athletic populations. For instance, Magee et al. (2025) employed ADP alongside DXA in NCAA Division I athletes, developing a novel estimation equation that significantly improved the accuracy of BF% prediction. Similarly, although conducted slightly earlier, Antonio et al. (2018) compared ADP, DXA, and bioimpedance spectroscopy (BIS) in collegiate football players and reported no significant differences in FM or BF% between the methods, supporting the validity of ADP in sport populations. In addition, Ahmadi et al. (2024) emphasized in their methodological review that both DXA and HW should continue to be regarded as gold-standard approaches for validating alternative techniques in athletes. Taken together, these findings illustrate that while DXA and BIA dominate current practice due to their accessibility, ability to provide regional estimates, and suitability for large-scale testing protocols, the unique precision of HW and ADP remains indispensable for the development of accurate, sport-specific regression equations and for validating indirect field methods.

In conclusion, while DXA and BIA dominate current sport-specific practice, the methodological rigor of HW and ADP underscores their continued importance for advancing BC research. Their precision, test–retest reliability, and long-standing status as reference methods make them indispensable not only for validating field techniques but also for developing sport- and population-specific regression equations. Looking ahead, integrating these densitometric approaches with multi-compartment models and sport-specific assessment will be crucial for minimizing measurement error, enhancing comparability across methods, and generating more robust insights into the unique BC demands of different athletic populations.

Conclusion and Discussion

The accuracy, reliability, and applicability of body composition (BC) assessment methods are of critical importance for both clinical health practice and sports sciences. Densitometric techniques, particularly within 4C models, hold special relevance as they provide values closest to invasive cadaver-based measurements. Regression equations derived from these reference methods enable the prediction of variables such as BF%—which are otherwise difficult and costly to measure—using simple anthropometric parameters. These predictions create an effective framework for evaluating physical fitness, athletic performance, and health status, identifying individual risks, and guiding training interventions. Looking ahead, the integration of additional biological, environmental, and genetic variables into BC assessment models could enhance both their accuracy and clinical applicability, supporting the development of more holistic evaluation systems. Such integrative approaches have the potential to improve early detection and prevention of obesity, malnutrition, and growth disorders while offering a robust framework for tracking developmental trajectories and optimizing individualized interventions across diverse populations.

The question of whether DXA should be considered a reference method or primarily a tool for assessing bone mass becomes particularly critical in specific populations where alterations in bone tissue are more pronounced. For example, in children and adolescents, rapid skeletal growth and mineralization processes necessitate precise evaluation of bone mass to ensure accurate BC estimates. Similarly, in older adults or individuals at risk of osteoporosis, bone demineralization and age-related changes in skeletal density highlight the importance of using DXA not only for fat and lean mass estimation but also for reliable assessment of bone mineral content. Thus, the role of DXA extends beyond its methodological function, serving as a crucial instrument in populations where bone tissue variability significantly influences BC outcomes (Gonera-Furman et al., 2022; Marra et al., 2019). Given that DXA bias is influenced by sex, size, fatness, and disease state (Williams et al., 2006), caution is warranted when using DXA alone for longitudinal tracking in patient populations or athletes undergoing significant body composition changes.

During life stages in which the density of FFM undergoes significant variation—such as infancy, childhood, adolescence, aging, and athletic participation—D_b becomes especially critical in the assessment of BC. Developmental differences between boys and girls, along with physiological and hormonal changes observed in women and elderly individuals, further emphasize the need for population-specific approaches. Therefore, the use of regression equations incorporating densitometry, or preferably regression models derived from at least 4C approaches, is essential (Wells & Fewtrell, 2006; Marin-Jimenez et al., 2022). HW and ADP are considered densitometry-based gold-standard methods and play an important role in enhancing the reliability of data obtained from 2C models. However, several methodological limitations must be acknowledged. For instance, during HW, the participant's ability to perform maneuvers such as maintaining proper breath control underwater is a critical determinant of measurement validity (Nana et al., 2015). In children, older adults, or other specific populations, variables such as the estimation of residual volume may introduce error. Moreover, the use of assumed values in ADP devices may not fully capture individual physiological variability, representing another limitation. Nevertheless, these methods consistently outperform other indirect techniques in terms of accuracy and reproducibility, providing high validity and strong test–retest reliability (Fields et al., 2002; Wells & Fewtrell, 2006). Morphological and developmental factors also have a direct impact on performance and must be considered when interpreting BC results. These considerations reinforce the need for population-specific prediction equations and underscore the importance of reference methods that minimize error and improve comparability across groups. Collectively, these efforts will contribute to more accurate, reproducible, and clinically meaningful BC assessment across populations. Consistent with this, a study on youth athletes reported that BF% and FFM estimates derived from anthropometric methods and multiple BIA equations differed significantly and could not be used interchangeably, further emphasizing the importance of population-specific validation in this age group (Harbili, 2008).

Future Directions

Looking ahead, advancing BC research will require addressing the following priorities:

1. *Standardization and Normative Data:* Re-evaluating the validity of existing methods across age, sex, and sport-specific populations, using normative centiles to improve comparability.
2. *Device and Software Consistency:* Addressing variability among DXA manufacturers and BOD POD software versions through mandatory reporting, calibration procedures, and inter-laboratory harmonization.
3. *Pre-Assessment Protocols:* Codifying standards for hydration, recent exercise, menstrual cycle phase, gastrointestinal contents, and environmental conditions to minimize measurement error.
4. *Longitudinal Sensitivity:* Establishing smallest detectable change and minimal clinically important difference thresholds for both athletes and patients to guide decision-making.
5. *Integration with Advanced Analytics:* Combining densitometry with 3C/4C models, total body water, and anthropometry, and leveraging AI-based predictive modeling to develop lower-burden, field-applicable solutions.
6. *Research Priorities:* Expanding studies on hydration-sensitive modeling for pediatric, elderly, and clinical cohorts; examining pharmacokinetic and pharmacodynamic interactions with BC; performing cost-effectiveness analyses; and developing robust data governance and radiation safety frameworks to facilitate broader clinical translation.

This integrated approach will not only enhance the accuracy of BC assessment but also strengthen its translational potential, supporting evidence-based decision-making in both healthcare and athletic performance optimization.

The accurate assessment of BC is essential not only for individual health management but also for the follow up and optimization of athletic performance (Fields et al., 2002; Wells & Fewtrell, 2006). However, the validity of these methods depends heavily on careful implementation, adherence to standardized protocols, and consideration of individual prerequisites such as age, sex, physiological status, and the ability to comply with specific testing procedures (Nana et al., 2015). When applied appropriately, HW and ADP provide valuable reference values that enhance the accuracy of regression equations and contribute to the development of more comprehensive multi-component models. Collectively, these measures will further strengthen the role of densitometry in clinical health applications, public health surveillance, and sports science.

Author Contributions:

This work was designed, conducted, and written in its entirety by the sole author, who takes full responsibility for the content of the manuscript

Table 1 Formulas for Estimating Body Fat Percentage from Body Density (D_b)

Population	**Ages**	**Equations**	References
Overall population 2C Model	Adults	$BF\% = [(4{,}95 / D_b) - 4{,}50] * 100$	Siri, 1961
Overall population 2C Model	Adults	$BF\% = [(4.570 / D_b) - 4.142] * 100$	Brozek, et al., 1963
Boys	9-11 ages	$BF\% = (5.30 / D_b - 4{,}89) * 100$	Lohman, 1986
Girls	13-15 ages	$BF\% = (5{,}12 / D_b - 4{,}69) * 100$	Lohman, 1986
Children	7-12 ages	$BF\% = 5{,}30 / D_b - 4{,}89$	Lohman et al., 1984
Boys and Girls	0- 1.99 ages	$BF\% = (\{585 - 4.7[age^*]^{0.5} / D_b) - \{550 - 5.1[age^*]^{0.5}\}$	Westrate & Deurenberg, 1989
Boys	2-18 ages	$BF\% = (\{562 - 4.2[age^{**} - 2]\} / D_b) - \{525 - 4.7[age^{**} - 2])$	Westrate & Deurenberg, 1989
Girls	2-10 ages	$BF\% = (\{562 - 1.1[age^{**} - 2]\} / D_b) - \{525 - 1.4[age^{**} - 2]\}$	Westrate & Deurenberg, 1989
Girls	10-18 ages	$VY \% = (\{553 - 7.3[age^{**} - 10]\} / D_b) - \{514 - 8.0[age^{**} - 10]\}$	Westrate & Deurenberg, 1989
2C Model (TBW) Overall population		$BF\% = [(BW - TBW / 0.732) / BW] * 100$	Siri, 1961
3C Model Overall population		$BF\% = [2.118 / D_{b(HW)} - 0.780 (TBW/BW) - 1.354] * 100$	Selinger, 1977
4C Model Overall population		$BF\% = [2.559 / D_{b(HW)} - 0.734(TBW/BW) + 0.983(TBBM/BW) - 1.841] * 100$	Heymsfield et al., 1990
Abbreviations: **BF%**: Body Fat Percentage; **BW**: Body Weight; **D_b**: Body Density (kg/m³); **TBW**: Total Body Water; **TBBM**: Total Body Bone Mass; **C**: Component; $D_{b(HW)}$: Body Density by Hydrostatic Weighing.			

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