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EDITORIAL

Evaluation of body composition: why and how?[1,2]

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Abstract Evaluation of human body composition *in vivo* remains a critical component in the assessment of nutritional status of an individual. Whereas traditional measurements of standing height and body weight provide information on body mass index and, hence, the risk of some chronic diseases, advanced technologies, such as dual X-ray absorptiometry, air displacement plethysmography and various forms of bioelectrical impedance analysis enable the determination of soft tissue composition (fat and lean) as well as bone. This review summarises the physical bases of these methods and critically evaluates their accuracy in observational and interventional studies. It also discusses a new approach, bioelectrical impedance vector analysis, which assesses the hydration status of an individual, and includes pertinent examples of its novel applications in clinical nutrition.

Keywords Dual X-ray absorptiometry · Body volume · Bioelectrical impedance · Bioelectrical impedance vector analysis · Hydration status

Introduction

Evaluation of human body composition *in vivo* requires the objective determination of various levels of the chemical composition of the body [1]. This challenge involves the precise and accurate measurement of the soft tissue composition (fat and fat-free or lean) and the bone mineral (mass and quality) of an individual. These measurements may be used to identify differences between groups (static appraisals) or to determine the responses to intervention or treatment (dynamic assessments) for an individual. The overall goals of body composition assessment *in vivo* for dieticians and clinical nutritionists are to quantitate body energy stores (fat or adipose tissue), structure (bone) and/or functional capacity (muscle mass or body cell mass). More than a century of research has produced diverse methods that utilise specific assumptions and models to accomplish these goals [2, 3].

This presentation concisely reviews and critically evaluates the use of some contemporary methods of evaluating human body composition *in vivo*. It emphasises dual X-ray absorptiometry (DXA), whole-body and segmental bioelectrical impedance techniques, and high-

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lights the novel application of bioelectrical impedance vector analysis (BIVA) to assess the hydration status of individuals in a variety of clinical conditions characterised by disturbances in water balance.

Dual X-ray absorptiometry (DXA)

This radiographic method utilises the graded attenuation of a beam of X-rays by bone, fat-free and bone-free tissue (lean soft tissue), and fat to estimate bone mineral and soft tissue composition. The beam of X-rays is pulsed at two discrete energy levels and administered in a posterior–anterior direction; the attenuation of the X-rays is measured with highly sensitive detectors positioned at a fixed distance from the scan bed and above the anterior surface of the body [4]. DXA overcomes the limitations of traditional methods such as underwater weighing, anthropometry and isotope dilution to estimate body composition because it is independent of assumptions of the constant chemical composition of the fat-free body, which is inherent in the two-component model of body composition [5]. Other beneficial characteristics are the rapidity of the measurement (5–15 min) and the capability to estimate regional and whole-body composition. Concurrent with the determination of soft tissue composition, DXA assesses bone mineral content and areal density. A practical limitation of DXA is exposure of the subject to ionising radiation (<5 μSv), which is low relative to daily radiation exposure (7 μSv) [6].

Figure 1 highlights the value of DXA in assessment of human body composition and shows the compositional variations between two adults with the same body mass index (BMI). Clear differences in the fat-free and bone-free mass (54 *vs.* 49.8 kg), fat mass (13 *vs.* 17.6 kg), fatness (19 *vs.* 25%) and bone mineral content (2696 *vs.* 2504 g) are evident in these images. This observation is consistent with previous findings that BMI is a non-specific indicator of body composition in healthy adults [7–9].

Alterations in body hydration confound DXA determinations of soft tissue composition. Studies of volunteers before and after ingestion of a standardised volume of water [10], voluntary dehydration [11] or renal dialysis [12] reveal that DXA identifies the increase or decrease in fluid volume as a change in body mass and lean soft tissue mass. Neither fat mass nor bone mineral content is significantly affected by alterations in fluid status up to 3% body weight. Thus, small but systematic and predictable errors in DXA estimates of soft tissue composition can arise with modest changes in hydration.

Body thickness is another factor moderating the validity of DXA determinations of soft tissue composition and bone. The accuracy of DXA determinations of body composition, compared with direct chemical analysis of animal carcasses, decreases significantly when body regions exceed 23 cm in anterior–posterior thickness [13].

The development of DXA for body composition assessment, particularly in multicentre trials, has evolved from pencil-beam to fan-beam instruments. Advantages of the fan-beam devices include a decrease in scan time and improved image resolution. An early validation trial of a fan-beam DXA with magnetic resonance imaging (MRI) found that DXA overestimated fat-free or lean soft tissue mass and underestimated fat mass [14]. A later investigation confirmed these findings and explained the discrepancy based on differences in the chemical and anatomical components measured with each technology

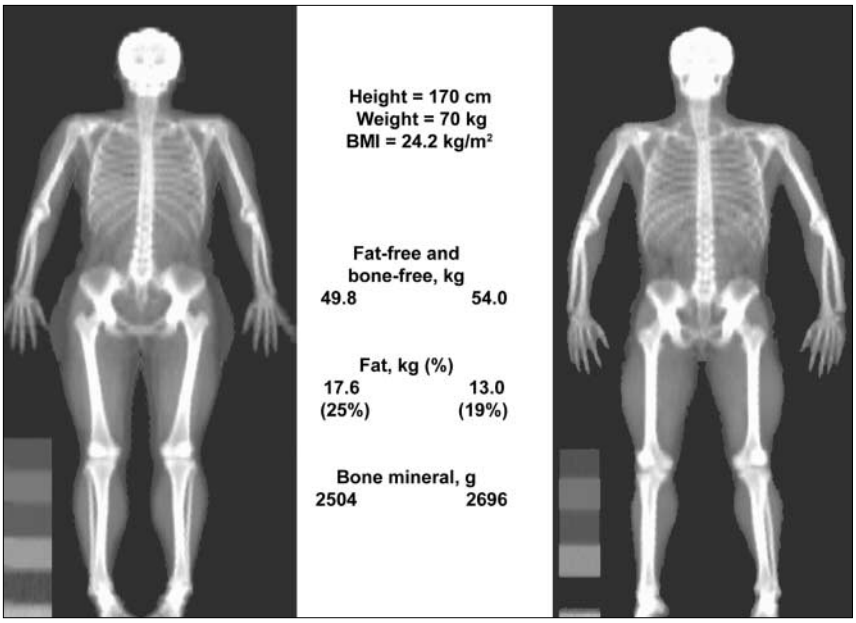

Fig. 1 Differences in the soft tissue composition and the bone mineral content of two adults with the same BMI

[15]. Lean soft tissue composition estimated with DXA includes not only skeletal muscle but also skin, connective tissues, protein matrix of adipose tissue and organs, whereas MRI only determines operator-delineated muscle mass and organs. Similarly, DXA-derived fat mass was less than the corresponding MRI-determined fat because DXA assesses chemical fat and MRI measures operator-defined adipose tissue including protein, mineral and water content.

Comparisons of DXA determinations of soft tissue composition with standard reference methods yield significant correlation coefficients ($r \geq 0.90$) but some discrepancies [16]. Pencil-beam and fan-beam devices produce modest differences in their estimates of body fatness (<4%) [17–19]. Also, differences in software programs to analyse DXA images produce divergent results (~2%) [20]. Thus, it is advised to use the same DXA instrument and analytical software version for any investigation, particularly longitudinal, multicentre interventional studies, and to include an appropriate anthropometric phantom (soft tissue or bone) for quality control and assurance [21].

Air displacement plethysmography (ADP)

Determination of body density, which is inversely related to body fatness, is an established measure in the assessment of human body composition [5, 22]. Although underwater or hydrostatic weighing is the traditional method for measuring body volume and calculating body density (density = weight/volume), this technique and its procedures can be problematic to perform for special populations including children, the elderly and the disabled. A safe and non-invasive alternative is ADP, whose popularity is growing. Body volume and density determined with ADP use a commercial device (Bod Pod, Life Measurement System Inc., Concord, CA) [23]. This technique requires subjects to sit in an enclosed test chamber, wear minimal clothing (e.g., tight-fitting swim suit), a swim cap, have minimal body hair, dry skin and hair, and perform specific breathing manoeuvres to estimate thoracic lung volume, which is used to calculate total body volume [24].

The reproducibility of ADP determinations of body fatness is very high. Mean within-day variability is 1.7–3.7% with one operator [25–28] but increases to ~5% with multiple operators [28]. Average between-day reproducibility over 3–7-day periods is 2–2.3% [28–30]. Thus, the variability of ADP determinations of body fatness exceeds the values (1–2%) reported for underwater weighing [31] and DXA [16].

Reports of the validity of ADP to assess body fatness in healthy subjects are not in agreement. Compared to underwater weighing, ADP either significantly overestimates [32–35] or underestimates [36–39] body fatness by 1–4% in adults. However, other reports indicate no difference between these methods [25, 27, 29, 30, 40]. A limited number of reports find that ADP either significantly overestimates [41] or underestimates [42] the body fatness of children and adolescents whereas other studies show no difference [30, 38].

Validation trials of ADP compared to DXA also describe inconsistent results. More reports find that ADP significantly underestimates [26, 29, 37, 39] than overestimates [33, 43, 44] body fatness in adults, with a growing number of reports indicating no significant difference between the methods [28, 30, 40, 45]. Among children, however, ADP, compared to DXA, significantly underestimates body fatness [41, 42].

Some factors contribute to the lack of consensus of the validity of ADP to estimate body fatness. They include the reliance on the two-component model of body composition (e.g., hydrodensitometry) that is prone to error associated with the between-subject variation in the chemical composition of the fat-free body [5, 22], and the use of different X-ray technologies and software analysis programs [16]. In the few studies that have utilised the four-component model to account for the between-subject differences in hydration and bone mineral density, the errors in estimation of body fatness are 2–3% [46].

Wearing excess clothing during the body volume determination can affect the validity of ADP to determine body density. Among lean adults, excess clothing (e.g., shorts, t-shirts, scrubs), as compared to swim suits, worn during the ADP results in a significant underestimation of body volume because the air in contact with the cloth remains isothermal as the pressure changes in the chamber. The more cloth worn, the larger the layer of isothermal air volume between the cloth and body surface. Because isothermal air is very compressible, body volume is underestimated, resulting in an overestimation of body density and decreased body fatness (2–5%) [32]. However, additional clothing (e.g., standardised cloth pants and short-sleeved tops) did not affect ADP estimates of body volume and composition in obese adults because the clothing fit tightly to body surfaces and minimised the isothermal layer as compared to lean individuals on whom the clothing fit loosely [44]. This finding has important practical implications for individuals with negative body image, such as overweight and obese adults and children.

Bioelectrical impedance analysis

Ionic solutions in the intracellular and extracellular spaces oppose the conduction of a low-level, radiofre-

quency, alternating current introduced into the body. This opposition, termed electrical impedance (Z), consists of resistance (R) and reactance (Xc) [47]. Investigators use four surface electrodes placed on the arm (wrist) and leg (ankle) with the subject supine and administer a low-level, alternating current at either single (50 kHz) or multiple frequencies (e.g., 1, 5, 50, 200, 500 and 1000 kHz), and then measure the whole-body Z, R and Xc if a phase-sensitive device is used. These variables are normalised for standing height (e.g., H^2/R) to control for differences in conductor length, and then combined with various physical and demographic variables (e.g., body weight, age, gender, etc) into regression models to predict total body water (TBW) or fat-free mass, assuming a constant hydration level (~73%) [48]. The consensus of evidence is that this 50-kHz, whole-body technique, as compared to reference methods, can lead to inaccurate predictions of body composition variables in some circumstances. Concerns include the use of sample-specific prediction models derived in groups of healthy individuals with relatively homogeneous composition can result in errors and wide ranges of estimates (e.g., large confidence intervals of prediction) when applied to individuals with different characteristics (e.g., body size, sex, ethnicity obesity, and hydration status) compared to the group in which the prediction model was developed [49, 50]. This single-frequency approach, however, yields useful body composition estimates that describe groups in epidemiological surveys [51, 52].

Another four-electrode method uses *a priori* selected pairs or ratios of impedance values derived at multiple frequencies in the prediction of fluid distribution [e.g., TBW, extra- and intracellular water (ECW and ICW)] and body composition of individuals with chronic diseases [53–55]. This approach assumes that low and high frequencies are needed to estimate ECW and TBW, respectively [56]. Overall, the use of multiple frequencies does not improve estimates of body fluid volumes as compared to the results obtained with single-frequency (50 kHz) impedance [57–61].

Another development is the use of mathematical modelling of multiple-frequency impedance data and mixture models of single cells in solution to develop equations to predict TBW and ECW [62]. This method uses empirically derived constants for intra- and extracellular resistivity for which there is wide discrepancy [63, 64]. This problem contributes to inconsistent findings in the application of bioelectrical impedance spectroscopy (BIS) in various clinical conditions (e.g., obesity, renal dialysis, pulmonary disease and muscle wasting). Some reports find that BIS improves accuracy [65, 66], has no improvement [67–69] or increases error [70, 71] as compared to other impedance approaches and tracer dilution reference methods. A recurrent pattern of BIS errors in the estimation of TBW and ECW water in samples of adolescents and adults [72, 73] has prompted a revision of the constants used in the resistivity calculation models. One successful attempt was to include BMI in the prediction models [74] to account for differences in body shape, which *per se* affects *Z* values independently of fluid distributions. Additional studies are needed to evaluate the validity of the inclusion of BMI in prediction of fluid distribution and body composition in healthy individuals and patients with altered fluid status and body composition.

Four-electrode, segmental bioelectrical impedance devices

A proposed, practical alternative to the whole-body, four-electrode method is segmental, four-electrode bioimpedance with contact (tactile) electrodes that measure impedance in the upper (arm-to-arm) or the lower (leg-to-leg) body of a standing subject [75–78]. The attraction of this segmental impedance technique is practicality and convenience for routine, personal monitoring of body composition outside of the laboratory because it eliminates the need for an operator of the device, the placement of adhesive electrodes on the wrist and ankle, and the wait for fluid equilibration with the subject in a supine position. The fundamental assumption of this technique is that conductor volume (e.g., ionic fluids and lean soft tissue) is equally distributed in the upper and lower body, and that segmental impedance is proportional to whole-body impedance [79, 80]. Although segmental and whole-body Z values are highly correlated, segmental impedance values are consistently different to whole-body determinations in adults [75, 76, 81] and children [82, 83].

Comparisons of body fatness values determined with segmental, four-electrode devices and DXA yield inconsistent results. A few reports show good correspondence of body fatness predictions with a foot-to-foot impedance device [75, 76] whereas another report [81] using a similar segmental device finds a significant underestimation of body fatness in men (1.7%) and women (2.6%). A hand-to-hand impedance instrument also significantly underestimates body fatness in men (2.3%) and women (6.3%) [81]. Interestingly, a regression model developed in an independent sample and containing only weight, stature, age and sex provides similar estimates of body fatness as DXA, but significantly overestimates body fatness compared to the segmental impedance devices [81]. A concern of these segmental devices is the validity of the proprietary prediction models and the importance of the measured impedance in the estimation of body fatness.

Eight-electrode, segmental bioelectrical impedance

This approach uses eight electrodes, four of which are embedded in the handles (thumb and palm) and another four in the foot scale pads (ball of foot and heel) of the device. This arrangement permits measurement of the impedance of the upper, lower and whole body at either single or multiple frequencies while the subject stands.

Validation trials of single-frequency (50 kHz), eight-electrode bioelectrical impedance devices find consistent errors. Studies of children and adults report a 2.6% overestimation of body fatness [84], 2.6% in lean young adults [85], 3.5% in normal-weight and 5% in overweight adults [86], obese women [87] and adults [88]. These findings suggest a trend of larger errors in estimation of body fatness with increasing obesity. Similarly, other validation trials using eight-electrode impedance instruments to estimate TBW and ECW report significant errors of 2–3 l in adults [89–91].

Other eight-electrode, segmental instruments introduce an alternating current (25 μA) at different frequencies (5, 50 and 250 kHz or 1, 5, 50, 250, 500 and 1000 kHz) into the body. The hypothetical advantage of this multiple frequency technology is to improve the estimation of fat-free mass, and hence body fatness, by determining ECW and ICW. A comparison of body fatness derived with these instruments finds similar values as determined by using the four-component model in men but a significant underestimation (~4%) in women [92]. Other studies report significant underestimation of body fatness (5%–6%), compared to DXA, in adults with a wide range of BMI levels [88, 93]. These findings suggest no improvement in the estimation of body fatness for individuals with eight-electrode, multiple-frequency, segmental impedance devices.

Bioelectrical impedance vector analysis (BIVA)

Altered hydration status or fluid imbalance is present in many physiological and disease states. Assessment of fluid components (e.g., TBW and ECW) requires tracer dilution methods that are impractical because of the need for trained personnel and costly equipment, the inability to provide real-time determinations and the lack of appropriate reference standards for an individual. Also, the use of prediction equations based on weight, height, age and sex, and bioelectrical impedance variables yields poor accuracy (>10%) for an individual assessment [52]. Thus, measurements of fluid volumes are very limited in clinical practice, and indicate the need to assess hydration as a practical alternative.

Whole-body Z, expressed as a vector, is a combination of R (opposition to the flow of an alternating current through intra- and extracellular ionic solutions) and Xc (capacitance produced by tissue interfaces and cell membranes) across tissues. The arc tangent of Xc/R is termed the phase angle (phase difference between voltage and current and determined principally by Xc). Interpretation of these variables suggests that the length of the Z vector is inversely related to fluid volume whereas the phase angle offers insight into the relative distribution of fluids (Fig. 2). Normalisation of Xc and R with the standing height (H) of an individual enables standardisation of these variables for differences in conductor length. This approach, which is termed bioelectrical impedance vector analysis (BIVA), allows for the evaluation of hydration and body composition without the need for prediction models [94].

Applications of BIVA focus on description of vector components in health and disease. Characterisation of a group (e.g., healthy or fluid overloaded) uses the mean group vector that is plotted on the R-Xc mean graph (Fig. 3) and the 95% percentile confidence interval, which is

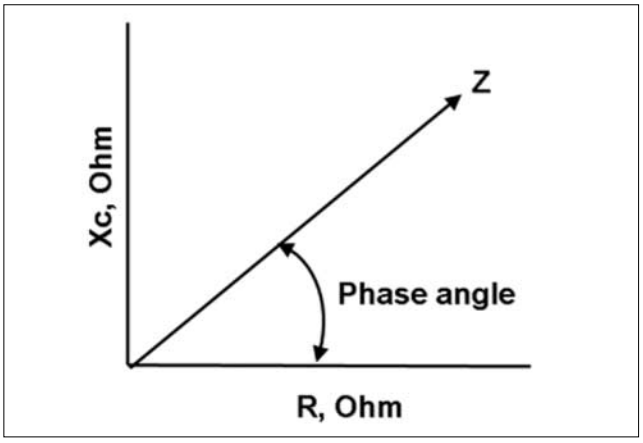

Fig. 2 Geometric components of the impedance (Z) vector including reactance (Xc), resistance (R) and the phase angle

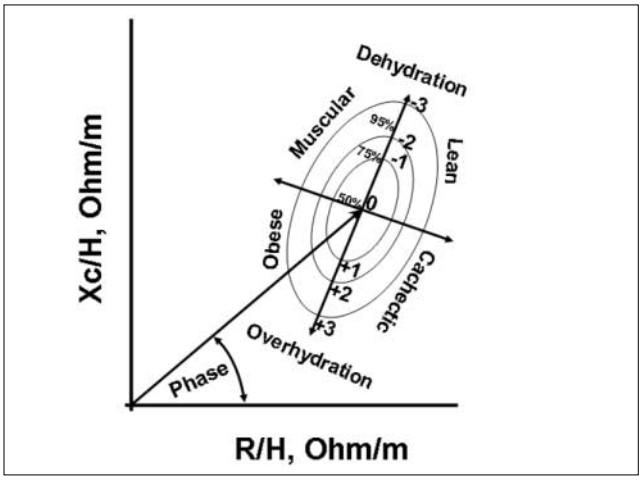

Fig. 3 Conceptual model of bioelectrical impedance vector patterns for clinical use (courtesy of A. Piccoli, University of Padua)

shown as an ellipse. This approach enables a comparison of vectors and tolerance ellipses of groups with different characteristics (e.g., sex, BMI, age, ethnic background, etc.) [95] and changes in these patterns in response to various treatments [96].

Clinical applications of BIVA, however, require a comparison of the affected group or individual vectors with a healthy reference population matched for sex. The mean reference vector is plotted with intervals or prediction ellipses that include 50, 75 and 95% bivariate percentiles [97]. Figure 3 illustrates the use of a general model of BIVA in assessing alterations in fluid status and body composition with the reference intervals derived in a sample of healthy men. For interpretation, the major axis indexes hydration status and the minor axis reflects tissue mass.

The focus of the vast majority of clinical applications of BIVA is characterisation of hydration status in different patient populations. Studies of patients with renal disease, compared to controls, show significantly shorter vectors with lower (smaller) phase angles that lengthen after dialysis [96]. Vectors of unstable (e.g., adverse outcomes), compared to stable, haemodialysis patients are longer with smaller phase angles; these differences between the patient groups persist after haemodialysis. Similar findings occur in peritoneal dialysis patients after fluid removal [98]. A fundamental outcome of these studies is the delineation of the 75% tolerance ellipse as the indicator of the boundary of normal tissue hydration; vectors outside the upper pole of the 75% ellipse indicate dehydration, whereas other vectors outside the 75% ellipse of the lower pole are characteristic of fluid overload or overhydration (Fig. 3).

Bioelectrical impedance vectors can also reflect impaired physiological function. In patients in an intensive care unit, central venous pressure (CVP) was inversely correlated with impedance measurements [99]. Regardless of sex, the vector of patients with low CVP was within the normal hydration range (<75% tolerance ellipse) and vectors from patients with high pressure were outside the 95% reference tolerance ellipse. Vectors from patients with medium CVP were distributed across the lower pole of the 75% ellipse. Thus, progressively increased CVP values are associated with shorter and down-sloping impedance vectors on the R-Xc graph. In contrast, dehydration induced with acute exposure to high altitude results in a significant lengthening of individual impedance vectors from the 50% ellipse at sea-level to the upper pole of the 75% percentile of the reference ellipse [100].

Regional measurements of impedance vectors are more sensitive than whole-body measurements in assessing localised fluid imbalance. Among patients with oedema subsequent to vascular surgery, vectors on the affected leg shortened significantly from pre-surgical condition with no change in the control or non-operated leg [101]. Although a similar response was found on the whole-body side with oedema compared to the control side, the magnitude was smaller. Similarly, BIVA determinations on the abdomen, compared to the limbs of patients undergoing peritoneal dialysis, were significantly more responsive to fluid loss [102].

There is increasing use of BIVA to monitor hydration changes during biological maturation. BIVA discriminates sexual maturity in females with a shorter impedance vector and increased phase angle in postmenarcheal compared to premenarcheal girls [103] and pre-pubescent children compared to adults [104]. In contrast, the vectors of older compared to younger elders were found in the lower right region of the 75% reference ellipse that is indicative of loss of body cell mass [105, 106].

A unique application of BIVA is the discrimination of fat and fluid changes in obesity. Compared to healthy adults with normal BMI (controls), obese individuals have shorter vectors and similar phase angles. However, obese adults with oedema show significantly shorter vector length than the normal weight and otherwise healthy obese adults, and significantly lower phase angle [107]. Vector lengthening occurs with fluid loss (3.2 kg) in the oedematous subjects whereas weight loss (9 kg) in only obese adults does not change vector length. Among healthy women with a wide range of BMI values, mean vector displacement shortens progressively with increasing BMI but phase angle remains constant [108]. The latter finding indicates increased soft tissue mass with normal hydration (e.g., ECW/TBW).

To date, the emphasis in the use of BIVA has been on characterisation of empirical changes in vector displacement with altered hydration states. There are few reports objectively validating BIVA. In response to haemodialysis, changes in the volume of fluid removed are significantly correlated with changes in vector components [96]. Additionally, measurements of TBW made before, during and after pregnancy are significantly correlated with vector length (r = –0.79). Also, changes in vector length are significantly related to changes in TBW (r = –0.599) [109]. Importantly, women whose individual vectors were outside the lower pole of the 75% reference ellipse had significantly greater increases in TBW during the third trimester (10 *vs.* 5 l) as compared to the other women whose vectors were within the 75% reference ellipse. These findings indicate the validity of BIVA to assess hydration and the sensitivity of this method to identify individuals with fluid overload.

With the increasing use of BIVA there is a need to standardise its application. A critical component is the

use of only phase-sensitive impedance instruments. Reports indicate significant errors in determination of Xc (10–12 ohm) and R (10 ohm) with impedance devices that do not determine Xc directly. Regular calibration of impedance instruments with appropriate resistors and capacitors is needed to assure quality control in studies using BIVA. There is also a need to operationally determine the optimal size of the surface electrodes because wide variance in Xc (20%) can occur with different surface area of the contact electrodes. Because Xc plays a critical role in establishing the position of the individual vector and the phase angle, technical factors that affect the validity of the Xc determination must be understood and controlled.

Conclusions

Measurement of human body composition is a fundamental component of nutritional assessment, with caution advised for the use of newer methods. DXA provides important information regarding soft tissue composition and bone (mass and quality). Practitioners should use a single technology (pencil or fan beam) with the most current software for image analysis. The attraction of bioelectrical impedance analysis should be tempered with the understanding that four-electrode, single-frequency applications are limited to epidemiological or observational studies. Multiple-frequency approaches, particularly impedance spectroscopy, are evolving and require additional validation before routine use can be recommended. Segmental, single- and multiple-frequency (four- and eight-electrode systems) devices yield questionable results and should be considered at a developmental stage. The new technique, BIVA, offers unique opportunities in clinical nutrition for practical assessment of hydration status and body cell mass without the limitation of reliance on assumptions associated with other methods.

Conflict of interest The authors declare that they have no conflict of interest related to the publication of this manuscript.

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