

PAPER

Body fat measurement in Indian men: comparison of three methods based on a two-compartment model

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Obesity is a major risk factor for diabetes and related disorders. The current classification of obesity is based on body mass index (BMI, kg/m²), which is a surrogate for the total body fat. Since the relationship between BMI and body fat varies in different populations, an independent validation of the BMI–body fat relationship in the population of interest is desirable.

OBJECTIVES: (1) To study the validity of field methods of measuring body fat (multiple skinfolds and bioimpedance) against a criterion method (deuterium dilution) and (2) To compare the prevalence of obesity (WHO 2000 criteria for BMI) with adiposity (body fat > 25%) in middle-aged Indian men in rural and urban Pune.

DESIGN: Community-based multistage stratified random sampling of middle-aged men from rural and urban Pune for study of body composition and cardiovascular risk. A third of these men, selected to represent wide BMI distribution, were studied for body fat measurements by specific methods.

SUBJECTS: A total of 141 healthy men, approximately similar number from rural, urban slums and middle class from Pune. They were 39.3 (± 6.2) y old and had a BMI of 21.9 (± 3.7) kg/m².

MEASUREMENTS: Anthropometry (height, weight and multiple skinfold thicknesses) by trained observers using standardised technique to calculate body fat by Durnin and Womersley's equation. Total body water and body fat by bioelectrical impedance analysis (BIA) and deuterium oxide dilution (D₂O).

RESULTS: Mean total body fat was 14.3 kg (23.0%) by anthropometry, 16.5 kg (26.0%) by BIA and 15.3 kg (24.6%) by D₂O method. Although there was a good correlation between fat estimation by three methods ($r = \sim 0.9$, $P < 0.001$ all), compared to D₂O method anthropometry underestimated body fat by 1.0 kg and BIA overestimated fat by 1.2 kg ($P < 0.001$ both). Using the standard cut-point of 25% body fat for 'adiposity' 29.5% rural, 46.0% slum and 75.0% middle class men were adipose. These proportions were considerably higher than the number of men who were 'preobese' (BMI ≥ 25–29.9 kg/m², 9.0% rural, 22.0% urban slums and 27.0% urban middle class) and 'obese' (BMI > 30 kg/m², 4.0% urban slums, none in rural and urban middle class).

CONCLUSION: We recommend that future studies assessing risk for chronic diseases in Indians should measure adiposity by anthropometry (multiple skinfolds) or BIA (calibrated for Indians) rather than relying only on BMI cut-points.

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Introduction

India is experiencing an epidemic of type 2 diabetes and related disorders.^{1–3} Obesity is a major risk factor for insulin resistance and type 2 diabetes. Body mass index (BMI, kg/m²) is the most commonly used measure of obesity.^{4–5} BMI is easy to measure in clinical and epidemiological studies

but it does not directly measure body fat. Prevalence of obesity in a population is determined as the proportion above a BMI cut-point. This is thought to represent number of individuals with excess amount of body fat.⁶ The relationship between BMI and total body fat differs in different populations.⁷ It appears that Indians have a different BMI–body fat relationship compared to Caucasians and African Americans, and that Indians are more adipose for a given BMI.^{8–15} Most of these findings are based on migrant Indians living in developed countries. We therefore studied the BMI–body fat relationship in rural and urban Indians in India.

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Intrinsic to the accurate characterization of the body fat–BMI relationship is the accurate estimation of total body fat. The easiest method to use in field situations is the model in which human body is divided into two compartments: fat and fat free mass (FFM).^{16,17} Two such methods are anthropometry (multiple skinfolds) and bioelectrical impedance analysis (BIA), which indirectly measure body density and body water respectively. However, these relatively easy to perform field methods require validation against a criterion method with minimal error.¹⁸ Deuterium dilution method (D₂O) is simple to perform in large population samples and we used this as the criterion method to compare anthropometric and BIA measurements of body fat in Indian middle-aged men.

Methods

This investigation is a part of the CRISIS study (Coronary Risk of Insulin Sensitivity in Indian Subjects), which investigated the relationship of body fat with insulin resistance and cardiovascular risk in middle-aged Indian men.¹⁹

Subjects

We aimed at studying ~150 apparently healthy men between 30 and 50 y of age from three residential areas in and around Pune (rural, urban slums and urban middle class). They were selected by multistage, stratified random sampling. Rural subjects were selected from two villages (Karandi and Dhamari) approximately 50 km from Pune city. Urban subjects were selected from 4/124 administrative wards in Pune city (2/55 slums and 2/69 urban middle class). Subjects were registered by a house-to-house survey and a total of 1222 men (30–50 y) were listed. We measured height and weight to define distribution of BMI in the whole group. After excluding known cases of diabetes, hypertension and cardiovascular disease we approached randomly selected subjects from each place with a view to study ~150 in each location. Finally, we studied 441 subjects (149 rural, 142 slums and 150 middle class). The participation rate was rural 86%, slums 79% and middle class 71%. D₂O studies were performed on a subset of 145 men from these 441 subjects by selecting a similar number of subjects from each tertile of the BMI of the whole group irrespective of their place of residence.

Measurements

Deuterated water method. Subjects reported at the Diabetes Research Centre, King Edward Memorial (KEM) Hospital, Pune, the evening before the study. They were given a standard dinner after which they rested.

At 5 h after dinner (about 0100 h) the subjects emptied their bladder completely to provide a basal urine sample. They then drank 75 mg per kg body weight of deuterated

water (Europa Scientific, Crewe, UK) from a sterile plastic container with the aid of a straw. This was followed by 3 g per kg body weight of plain water using the same straw and container.

Urine samples were collected every hour between 4 and 7 h after drinking deuterated water. Weight measured after the last urine sample was used for all calculations. Samples were frozen (at –70 °C) until transported to St John's Hospital, Bangalore, for further analysis. Deuterium enrichment was analysed by zinc reduction followed by dual-inlet mass-spectroscopy (Europa Scientific, Crewe, UK) as recommended by the IDECG²⁰ and as described earlier.²¹ Each sample was analysed in duplicate and the mean was used for analysis. Repeated analysis for natural background samples gave a cv (delta vs Standard Mean Ocean Water, SMOW) equal to 0.02%. With high enrichment samples using IAEA standard no 302 (enrichment equal to 500 vs SMOW), cv was equal to 0.22%.

The sample dose of deuterated water was also analysed to ascertain its enrichment and this was adjusted in the calculation of the total body water (TBW). The calculated value was downadjusted by 4% for hydrogen exchange to give the true TBW.²² FFM was calculated using a hydration constant of 0.732²³ and the difference between the body weight and FFM was taken as total body fat.

Anthropometry. Three trained observers performed all measurements. Height was measured to the nearest 0.1 cm using a wall fixed stadiometer (CMS Instruments, London, UK) and body weight was recorded to the nearest 0.1 kg using a portable scale (Soehnle, Waagen GmbH, Germany). The biceps, triceps, subscapular and suprailiac skinfold thicknesses were measured on the nondominant side of the body using Harpenden skinfold callipers (CMS Instruments, London, UK). The difference in measurement of skinfolds between observers ranged from –0.98 to 0.68 mm, resulting in an interobserver coefficient of variation <6%.

Durnin and Womersley's equation²⁴ was used to calculate body density from the sum of skinfold thicknesses. Siri's equation²⁵ was used to calculate body fat percent from body density. The difference between body weight and body fat was taken as FFM.

Bioelectrical impedance. The measurement was carried out using MultiScan 5000 (Bodystat Ltd, Isle of Man, UK) according to the recommendations in the NIH Technology Assessment Statement.²⁶ A specific Bodystat calibrator (500 ohms) was used daily to confirm the reproducibility of the measurement (coefficient of variation <0.2%). BIA test was carried out in the morning (fasting) after the subject had emptied bladder. For each subject, both the impedance value (50 kHz) and TBW calculated by the manufacturer's software were recorded. The manufacturer's software calculates TBW using an equation generated on 236 European Caucasian males with a wide range in age, height and body weight (information provided by manufacturer).

The study protocol was approved by the ethical committee of the KEM Hospital Research Centre, Pune and by the local community leaders in the three study areas. Each subject signed informed consent.

Statistical analysis. Data are presented as mean, standard deviation (s.d.), and range. Statistical significance of the difference between measurements obtained by three methods was tested by the paired *t*-test. Difference between group means was tested by the independent *t*-test. The anthropometric and BIA measurements of body fat were correlated with the criterion method (D₂O) by Pearson's method. The bias ($[(\text{observed value} - \text{criterion value}) / \text{criterion value}] \times 100$) and limits of agreement (mean difference ± 2 s.d.) in relation to the criterion method were assessed by Bland-Altman method.²⁷ We used multiple linear regression analysis to study the relationship between body fat measurement (D₂O method) and its determinants (impedance value and anthropometric measurements). All statistical analysis were performed using Statistical Package for Social Sciences (SPSS) for Windows (version 10.0).

Results

Of 145 subjects who participated in the study, three subjects who had low impedance values and therefore negative body fat mass were excluded from the analysis. These subjects were not significantly different from the whole group with respect to anthropometry and biochemistry and we could not find any methodological explanation for the unusually low readings. One subject had a very low body fat by D₂O method (1.4 kg, 2.6% body fat) probably because of technical error and was also excluded. The following analysis is therefore on 141 subjects.

There was no significant difference in the anthropometric characteristics of the parent group ($n = 441$) and the study group ($n = 141$). In the study group the range of heights and weights of the subjects were 149.8–180.7 cm and 37.8–100.6 kg respectively, giving a BMI range of 15.1–34.6 kg/m² (Table 1).

TBW values obtained by the D₂O technique (criterion method) ranged from 23.4 to 48.7 kg, FFM values ranged from 32.0 to 66.5 kg and body fat from 3.8 to 34.1 kg. This represented 9.2–40.4% of body weight. The anthropometric method estimated body densities (1.02–1.07 kg/L) giving total body fat 4.4–31.7 kg, which represented 11.3–36.2% of body weight. In the BIA method the primary estimate was TBW (using software) which ranged from 25.0–42.8 kg, giving FFM of 34.1–58.4 kg and total body fat of 1.0–42.2 kg (2.8–43.2% of body weight). The anthropometric and BIA methods correlated strongly with the D₂O method ($r = 0.92$, $P < 0.001$, both) (Figure 1a and b). However, there were small but significant differences between the values measured by these methods. To assess the agreement between the D₂O, anthropometry and

Table 1 Anthropometric and body composition parameters in the study group ($n = 141$)

Parameter	Mean (s.d.)	Range (minimum–maximum)
Age (y)	39.3 (6.2)	29.0–52.0
Height (cm)	165.5 (6.1)	149.8–180.7
Weight (kg)	60.0 (10.8)	37.8–100.6
BMI (kg/m ²)	21.9 (3.7)	15.1–34.6
<i>TBW</i>		
Anthropometry (kg)	33.5 (4.3)	24.3–52.7
BIA (kg)	31.8 (2.8)	25.0–42.8
D ₂ O (kg)	32.7 (4.3)	23.4–48.7
<i>FFM</i>		
Anthropometry (kg)	45.7 (5.8)	33.3–72.1
BIA (kg)	43.5 (3.8)	34.1–58.4
D ₂ O (kg)	44.7 (5.8)	32.0–66.5
<i>Body fat</i>		
Anthropometry (kg)	14.3 (5.9)*	4.4–31.7
BIA (kg)	16.5 (8.1)*	1.0–42.2
D ₂ O (kg)	15.3 (6.4)	3.8–34.1
Anthropometry (%)	23.0 (6.2)*	11.3–36.2
BIA (%)	26.0 (9.3)*	2.8–43.2
D ₂ O (%)	24.6 (7.0)	9.2–40.4

*Significantly different from D₂O, $P < 0.001$.

BIA methods we used Bland and Altman technique. The difference from the D₂O method was plotted against the mean of the two methods and the limits of agreement (2 s.d.) between the methods were calculated (Figure 2a and b).

When compared to D₂O method anthropometric method gave a mean fat mass difference (D₂O—Anthropometry) of 1.0 (± 2.5) kg (Figure 2a) while the BIA method gave a mean fat mass difference (D₂O—BIA) of -1.2 (± 3.3) kg (Figure 2b) (bias = -6.5 and 7.8% , respectively). Both of these mean differences were significantly different from zero ($P < 0.001$).

For anthropometry, the difference (D₂O—anthropometry) has no significant relationship with the average value (Figure 2a). However, for BIA the difference (D₂O—BIA) was inversely related to the average value ($r = -0.50$, $P < 0.001$). Thus, at lower values BIA underestimated and at higher values it overestimated the fat mass (Figure 2b).

The predictive equation derived from this data set for body fat (D₂O) is:

$$\text{Fat (kg)} = -8.021 + 0.472[\text{height}^2/\text{impedance}]$$

with $r^2 = 0.31$, $P < 0.001$, standard error of estimate 5.4 kg. Including body weight in the equation improved the prediction:

$$\text{Fat (kg)} = -12.297 - 0.287[\text{height}^2/\text{impedance}] + 0.694 \times [\text{weight}]$$

with $r^2 = 0.82$, $P < 0.001$, standard error of estimate 2.74 kg. The latter predictive equation was internally validated by dividing the whole group randomly into two equal-sized groups and inspecting the distribution of residuals

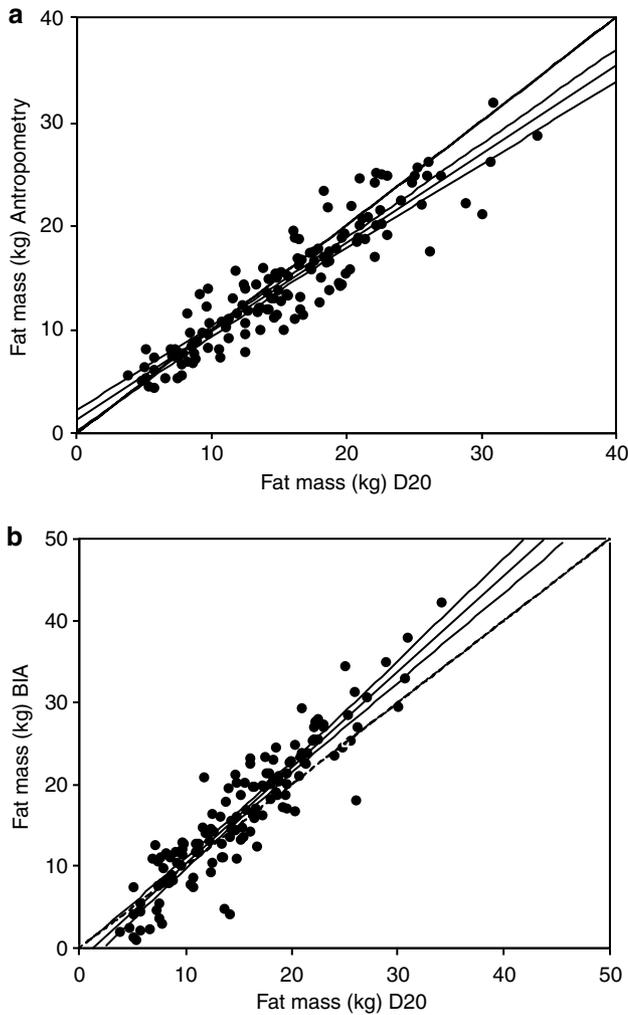


Figure 1 (a) Shows relationship between body fat mass by criterion (D₂O) method and by anthropometry ($r=0.9$, $P<0.001$). Dotted line is the line of no difference. Regression line and confidence interval shown. (b) Shows relationship between body fat mass by criterion (D₂O) method and BIA ($r=0.9$, $P<0.001$). Dotted line is the line of no difference. Regression line and confidence interval shown.

(mean = 0.00, s.d. ± 2.7) which is a normal distribution emphasizing the accuracy of the equation for this population. Total body fat using this equation ranged from 3.7 to 34.7 kg (9.7–35.1% of body weight). There was a strong correlation between fat mass obtained from manufacturer's equation and the new predictive equation ($r=0.92$, $P<0.000$), however, the software significantly overestimates the fat mass by 1.2 kg ($P=0.000$) with limits of agreement -3.3 to 5.9 kg.

Body mass index and body fat percent

We compared the relationship between body fat percent (D₂O, anthropometry and BIA methods) and the BMI. The body fat percent calculated by each method correlated

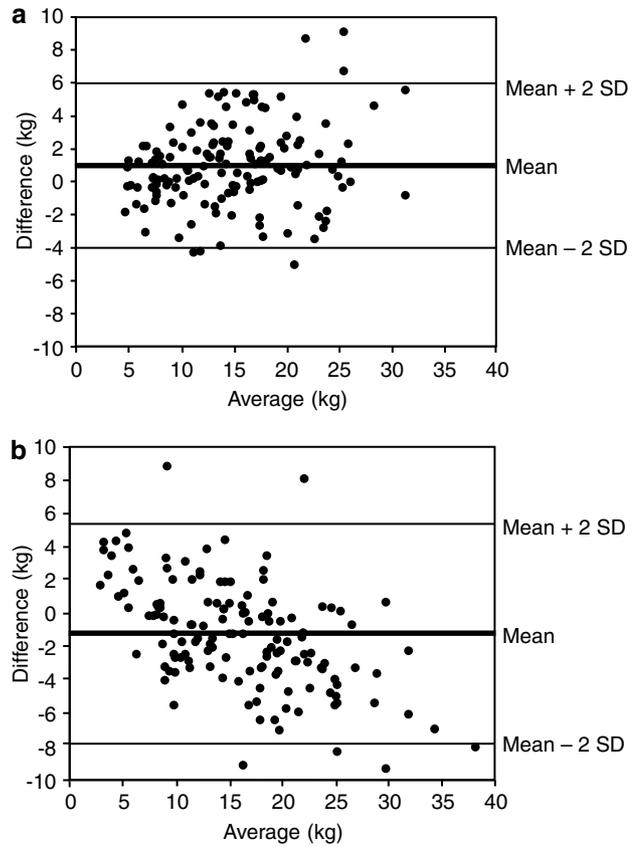


Figure 2 (a) Shows comparison of fat mass by the criterion (D₂O) method and anthropometry (Bland and Altman Plot). The limits of agreement between D₂O method and anthropometric measurements of body fat were (1 ± 5) kg. There was no systematic bias. (b) Shows comparison of fat mass between criterion (D₂O) method and BIA (Bland and Altman Plot). The limits of agreement between D₂O method and BIA measurements of body fat were (-1.2 ± 6.6) kg. BIA overestimated at higher body fat and underestimated at lower body fat.

strongly with the BMI ($r=0.76$, $P<0.001$ for D₂O method, $r=0.86$, $P<0.001$ for anthropometry method and $r=0.89$, $P<0.001$ for BIA method) (Figure 3a, b and c). For the D₂O method this is represented by the equation:

$$\text{Fat (kg)} = -7.085 + 1.441 \times \text{BMI}.$$

The figure shows that the equivalent BMI for a body fat of 25% by D₂O method is 22.1 kg/m², by anthropometric method it is 22.9 kg/m² and by BIA method 21.6 kg/m². In addition, the number of subjects with body fat above 25% ('adipose') was compared with the number of subjects with BMI above 25 kg/m² ('preobese' and 'obese'). In all, 52% of the subjects had body fat >25% (D₂O method), 40.4% (anthropometry method) and 57.4% (BIA method) while 19.1% had a BMI ≥ 25 kg/m². The figure also shows the number of individuals misclassified as 'normal' by BMI method in comparison to the three methods of body fat measurement; this number was 46 (32.6%) for D₂O method,

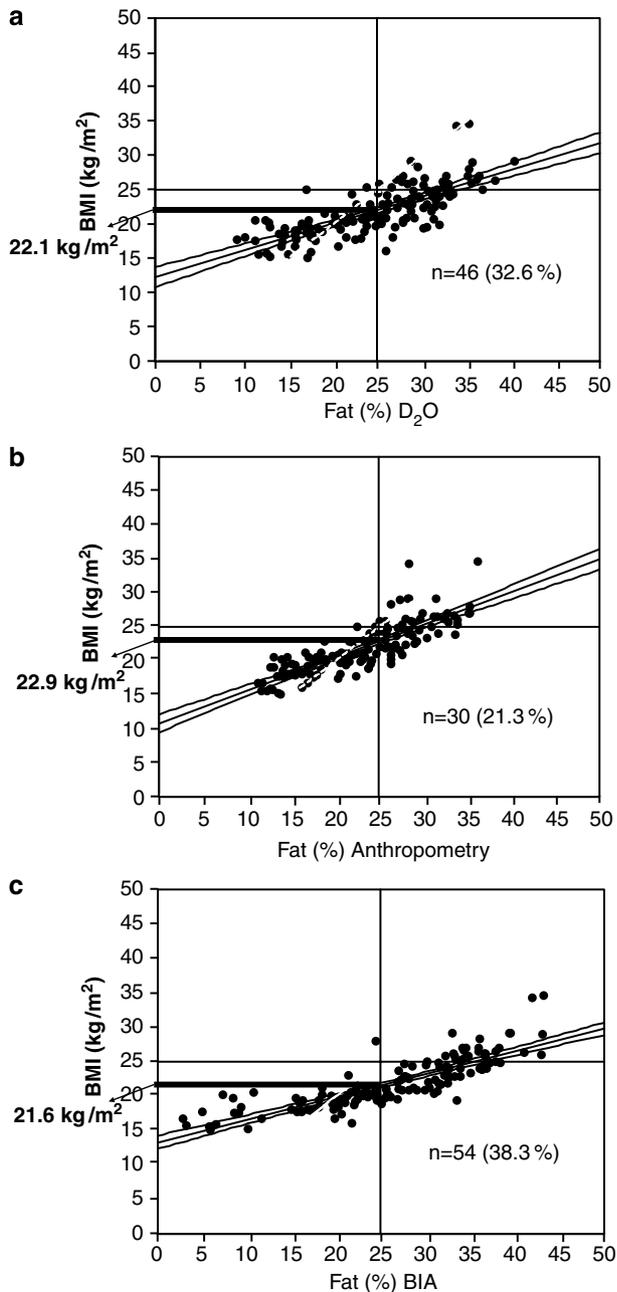


Figure 3 (a) Shows the comparison between body fat% (D₂O method) and BMI. Twenty five percent body fat ('adiposity') by D₂O method is equivalent to a BMI cut-point of 22.1 kg/m² and 32.6% individuals are misclassified as 'normal' by BMI compared to D₂O method. Regression line and its 95% confidence interval shown. (b) Shows the comparison between body fat% (anthropometry, Durnin and Womersley's equation) method and BMI. Twenty five percent body fat (adiposity) by anthropometry (Durnin and Womersley's equation) method is equivalent to a BMI cut-point of 22.9 kg/m² and 21.3% individuals are misclassified as 'normal' by BMI compared to anthropometry method. Regression line and its 95% confidence interval shown. (c) Shows the comparison between body fat% (BIA, MultiScan 5000) method and BMI. Twenty five percent body fat (adiposity) by BIA (MultiScan 5000) method is equivalent to a BMI cut-point of 21.6 kg/m² and 38.3% individuals are misclassified as 'normal' by BMI compared to BIA method. Regression line and its 95% confidence interval shown.

30 (21.3%) for anthropometric method and 54 (38.3%) for BIA method.

Discussion

To the best of our knowledge this is the largest population-based study comparing body composition methods in India. This paper reports results in middle-aged men. We found that in middle-aged Indian men, body fat measured by the two-field methods, anthropometry (Durnin and Womersley's equation) and bioelectrical impedance (MultiScan 5000, Bodystat) correlates strongly with that measured by the criterion method (D₂O). However, anthropometric method significantly underestimated and bioimpedance overestimated body fat compared to the D₂O method. There was no systematic bias in anthropometric method but bioimpedance underestimated at lower and overestimated at higher degrees of adiposity.

There is little good quality data in Indians of body fat measured by appropriate techniques and only limited information on relation between BMI and body fat.²¹ Most of the studies are in Indians abroad,¹⁰⁻¹² the only Indian study reporting BIA measurements in relation to cardiovascular risk factors used unvalidated manufacturer's equation.²⁸ Our subjects had a large range of BMI and body fat, allowing for a better determination of a prediction equation for body fat. It would be ideal to use four-compartment model as the criterion method with a higher degree of precision but it was not possible because of lack of appropriate technology and financial constraints. We used a criterion method that would have a precision of about 1 kg (in a 50 kg man, with 15% fat, and with 30 l TBW), calculated by propagation of error with a 2% and 0.1 kg precision of measurement of TBW and weight respectively.²⁹ D₂O method also has the advantage that it is relatively easy to perform in large number of subjects. However, a significant limitation of the deuterium dilution method is that it assumes similar hydration of the FFM in all individuals. Variation in the hydration of the FFM could occur due to age, as well as due to interindividual variation. In one report,¹⁸ this variation was about 3%, which would translate into an error of similar magnitude in determination of the FFM.

Despite a highly significant correlation between the criterion method and the skinfold and BIA measurement of body fat ($r \sim 0.9$) there are significant limitations to interpretation of skinfold and BIA results. Mean body fat measurements for the group by the three methods were similar but the limits of agreement between the criterion method and the two-field methods were rather large (for anthropometry -4.0 to 6.0 kg and for BIA -7.8 to 5.4 kg). This implies that skinfold and bioimpedance fat measurements for an individual will have large limits of confidence and this fact should influence the interpretation of results. On a population basis there was no systematic difference in the body fat measurement by skinfolds method in relation to D₂O measurements, proportional to the amount of body fat.

This suggests that skinfold fat measurement by Durnin and Womersley's equation may be used in Indians without any change in the prediction equation. This is welcome news for epidemiological studies, which are able to use only this method of body fat measurement. We had not expected such a close relationship because Indians have a different distribution of subcutaneous fat and visceral fat compared to white Caucasians in whom the original equation was defined.^{10–11,30}

On the other hand, BIA measurement showed a systematic difference in body fat measurement compared to D₂O method proportional to the amount of body fat, underestimating it at lower and overestimating it at higher levels. This could exaggerate the prevalence estimates of adiposity in an adipose population, viz. urban Indians. Given the large limits of agreement between D₂O and BIA methods an individual result should also be viewed with caution. There is therefore a need to devise new age- and gender-based Indian equations; this has been done for middle-aged Indian men in the present study. Our findings will have significant impact on the use and interpretation of results of portable BIA machines in India, either in clinical practice, gymnasiums offering 'slimming' programs or in research institutions.

Finally, we compared the number of individuals who were classified 'preobese' and 'obese' by the currently used WHO cut-points for BMI⁴ and the number who have body fat percent that was comparable to that predicted for Europeans at these BMI cut-points by an equation based on BMI, gender and age.³¹ The point estimate of body fat was made for men with a BMI of 25 kg/m² and an age of 45 y, and rounded off to the nearest half decile, which worked out to a body fat of 25%. In our study, 19.7% of middle-aged men were 'preobese' and 'obese' but 49.3% were adipose (>25% body fat by D₂O method). Thus, WHO BMI cut-points substantially underestimate adiposity in Indians. BMI cut-points are useful as population risk indicators for mortality as well as morbidity, the major component of which is the risk of type 2 diabetes. Two recent reports from India provide some support to our argument. Ten-year incidence risk of hyperglycaemia (impaired glucose tolerance + diabetes mellitus) was 2.4 times higher in middle-aged normal glucose tolerant Indians with BMI >23 kg/m² compared to those with a lower BMI.³² Similarly, in a large multi-centre cross-sectional study of diabetes prevalence in India, BMI of >23 kg/m² significantly predicted diabetes.³³ Similar observations in other Asians led to a recent WHO recommendation to create a lower BMI cut-point of 23 kg/m² for 'public health action' in Asians.³⁴ The next logical step in this process is to collect prospective morbidity and mortality data in relation to BMI in India and other Asian countries.

In summary, we compared two field methods of body fat measurement with a criterion (D₂O) method. Multiple skinfold measurements and use of Durnin and Womersley's equation slightly underestimated while a commercial BIA machine (MultiScan 5000, Bodystat) overestimated body fat

in middle-aged Indian men. BIA body fat measurements were also biased such that they underestimated at lower end and overestimated at upper end. We provide a new prediction equation to calculate body fat from bioelectrical impedance measurements in Indians. We recommend that future studies of body fat ('adiposity') in Indians should measure multiple skinfolds or use a specifically calibrated bioelectrical impedance machine rather than rely solely on BMI cut-points of obesity.

Conflicts

Dr CS Yajnik and Dr AV Kurpad were invited experts for the WHO Expert Consultation on Appropriate BMI cut-points for Asians, Singapore, 2002.

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