TOTAL BODY WATER MEASUREMENTS BY DEUTERIUM DILUTION IN ADULT INDIAN MALES AND FEMALES

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Abstract : Total Body Water (TBW) was measured in a group of 20 healthy adult Indian men and 10 women by the deuterated water dilution technique and their body composition was determined by applying a hydration factor of 0.7194 for fat free mass (FFM). The TBW in the male subjects whose mean body weight was 49.8 ± 6.7 kg, was $60.6 \pm 3.2\%$ of body weight (range 55.8%– 65.4%), from which a FFM of 41.9 ± 6.1 kg (range 31.8 kg–51.3 kg) was obtained. Total body water in the group of 10 female subjects whose mean body weight was 42.7 ± 4.9 kg, was 57.0 ± 4.5 % of body weight (range 52.5%– 64.2%) from which a FFM of 34.0 ± 5.1 kg (range 28.4 kg–39.4 kg) was obtained.

Key words: deuterium dilution   total body water

INTRODUCTION

Water is a major component of the human body and is broadly partitioned into an intracellular and an extracellular compartment (1). The most accurate method of human *in vivo* measurement of TBW is the tracer dilution technique which commonly uses deuterium (2,3,4) and tritium (5,6). Estimates of TBW may be derived as a percentage of the body weight, but vary in the literature from 52% to 64% for a whole group with an individual range of variation between 52% to 72% (7,8). The application of these estimates, without validation, to populations who tend to be underweight (such as those in India) may not be sound, as these estimates have been determined in predominantly Caucasian populations.

Accurate estimates of total body water by deuterium dilution can also be employed to obtain estimates of FFM, on the premise that fat mass is anhydrous and that all the water, is present in the FFM. Therefore, a hydration factor (9) may be used to derive values for FFM from the TBW. From this, it is possible to get estimates of the fat mass, and therefore describe the body in terms of 2 compartments: the fat mass and the FFM.

The aims of this study were twofold; Firstly, the study aimed at using the deuterium dilution technique to measure total body water in adult Indian men and women subjects. Secondly, the study aimed to describe the body composition of these subjects in terms of a two compartment model by applying a hydration factor of 0.7194 for the FFM.

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METHODS

Twentysix adult male and 11 adult female subjects in the age group of 20-40 years were recruited for the study. A brief medical history was taken and medical examination was carried out before the experimental protocol. Their weights were measured to the nearest 0.1 kg, and their heights to the nearest 0.1 cm. Ethical approval for the study was obtained from the Ethical Committee of the College, and all the subjects gave informed consent.

Of the 26 male subjects recruited for the study, 6 subjects drank water after they were dosed with deuterium thus invalidating their study. Therefore, data from 20 male subjects was used in the analysis. Data from 10 female subjects was also included in the analysis.

The experimental protocol for the male subjects commenced at 0100 hours during an overnight stay in laboratory. The protocol for the female subjects commenced at 0600 hours as they were unable to spend the night in the laboratory due to cultural reasons. The male subjects reported to the laboratory at 2100 hours on the previous night of the experiment and were housed in a metabolic ward having an ambient temperature of $26.0 \pm 2.0°C$. Each subject was given 200 ml of water to prevent thirst through the night following which no further water or food intake was allowed for the duration of the experiment. The male subjects were woken up and a basal sample of urine collected. After complete evacuation of the bladder, each subject orally consumed deuterium oxide (D_2O, 99.9%, Europa Scientific, Crewe, UK) in the dose of 40 mg/ kg body weight, from sterile plastic containers with the aid of a straw followed by 50 ml of distilled water using the same straw. The subjects were made to gargle with the distilled water before swallowing to wash down possible remnants of the isotope in the mouth.

The container was tightly sealed with the straw crushed within it and the post dose weight of the container recorded, to calculate the exact amount of isotope consumed. Urine samples were collected hourly from the 4th hour after dosing until the 8th hour, in new plastic containers and stored at 4°C. An identical experimental protocol was followed for the female subjects at 0600 hours.

The urine samples were analysed for deuterium using a dual inlet Mass Spectrometer (Europa Scientific, Crewe, UK). Samples were prepared using the Zinc reduction technique as recommended by the IDECG 1990 (10). The Mass Spectrometer consisted of a magnetic sector instrument in which the flight tube passed in a curve near a permanent magnet. After sample preparation gas samples were admitted into the ion source by means of a capillary leak. Since this was a dual inlet system, pressures on both sides of the inlet were controlled by a system of bellows. The gas molecules were ionised by electrons from the ion source and propelled by a positive potential into a magnetic field where they were resolved by the magnet into separate beams depending on their masses and admitted into separate collectors, by which their amplitudes were determined. The dual inlet system allowed for reference and sample gases to be admitted alternatingly into the source. A nearly simultaneous measurement of sample and reference (SMOW) hydrogen was obtained thereby eliminating errors of system drift.

Samples for each time point were analysed in duplicate. The post dose values for each time point were plotted to establish the 'plateau' of enrichment and the mean of these values (E_g) was used for calculation. The increase in enrichment was calculated by taking the arithmetic difference between post dose enrichment and the predose basal enrichment (E_p). The TBW was calculated by the formula:

$$TBW \ (kg) = \frac{Dose \ of \ D_2O \ administered \ (gm)}{Increase \ in \ enrichment \ (E_s - E_p)}$$

The TBW (kg) was converted into FFM by multiplying it by a factor of 0.7194 (hydration factor of FFM (11)). The fat mass was calculated as the difference between the body weight and the FFM.

Results were expressed as Mean ± SD. Values of TBW and FFM were regressed on variables of body weight, height and age for each of the two .groups. Statistical analysis for differences between groups was carried out by the Students independent 't' test. Regression analysis and correlation coeffecients were obtained by the Pearson's Product Moment correlation method. Results were considered significant if P < 0.05.

RESULTS

The male subjects had a mean age of 24.9 ± 3.5 years and weighed 49.8 ± 6.8 kg. Their mean height was 164.1 ± 5.3 cm which gave a Body Mass Index (BMI, Weight/Height^2, kg, M^{-2}. The female subjects had a mean age of 27.1 ± 6.0 years and weighed 42.7 ± 4.9 kg. The mean height was 153.5 ± 7.4 cm which gave a BMI of 18.2 ± 1.9 kg, M^{-2}

In the males, the TBW was 29.8 ± 3.5 kg, which was 60.6 ± 3.2% of the body weight (range 55.8%–65.4%) This gave an FFM of 41.9 ± 6.1 kg (using a hydration factor of 0.7194), and hence % Fat from body weight which was 15.8 ± 4.5%. In the female group, TBW was 24.4 ± 3.6 kg, which was 57.0 ± 4.5% (range 52.5%–64.2%) of the body weight. This gave an FFM of 34.0 ± 5.1 kg (using a hydration factor of 0.7194), and a % Fat of 19.7 ± 6.6.

TABLE I : Total body water (TBW) and Fat Free Mass (FFM) in adult Indian males and females.

	Males n = 20	Females n = 10
TBW (kg)	29.8 ± 3.5	24.5 ± 3.6*
FFM (kg)	41.9 ± 6.0	34.0 ± 5.1*

Results are Mean ± S.D.

FFM : Fat Free Mass by deuterium dilution
TBW : Total Body Water by deuterium dilution

P = *, if < 0.05, for comparisons between groups.

All the values for the females were significantly different from those of the male group. The TBW and FFM for both sexes are given in Table I. The relationship of the parameters of body weight, height and age and the obtained values of TBW and FFM for both the sexes are shown in Table II.

TABLE II : (Pearson's Product Moment) Correlation coeffecient and relationship of Total Body Water (TBW) with bodyweight (kg), height (cm) and age (years) for male and female subjects.

	r	r^2	Slope	Intercept	Significance of the relationship
Males					
Weight	0.79	0.63	0.53	3.77	*
Height	0.46	0.21	0.36	−29.74	*
Age	0.31	0.09	0.39	20.12	NS
Females					
Weight	0.81	0.67	0.59	−0.42	*
Height	0.51	0.26	0.25	−14.13	NS
Age	0.10	0.01	−0.06	26.31	NS

P = *, if < 0.05.

There was an expected significant correlation of TBW with weight in the male and female groups. Additionally, there was a weak correlation with height in the male group. All other relationships were not significant. The correlations of FFM with the anthropometric parameters showed the same relationship with the anthropometric variables; this was expected as FFM was a function of the TBW.

DISCUSSION

The mean BMI of the male and female subjects was 18.5 ± 2.0 kg M^{-2} and 18.2 ± 1.9 kg M^{-2} respectively. This study demonstrates that isotopically measured TBW as a percentage of body weight in healthy, but low BMI Indians is similar to those found in other populations (12), with values ranging from 55.8 to 65.4% in males and 52.5% to 64.2% in females. The variation is also similar to that found in earlier reports (8, 12) using the same techniques.

A number of substances, including deuterium, tritium, antipyrine, ethanol, urea and thiourea have been used to determine TBW. Tritium accurately measures TBW but is radioactive and also offers no great advantage in the accuracy of measurement of body water when compared to deuterium

(13). Moreover, it cannot be used in all groups of populations, such as pregnant women, children and medically ill patients. Antipyrine has been used (13,14), but has the disadvantages of being an invasive method and having a short half life which requires a rapid sampling rate (14). Ethanol has also been used (15,16) to determine the TBW, but has the disadvantage of a high level of dosing, non steady state kinetics, reabsorption from the bladder and gastrointestinal toxicity (15). These factors may account for the lower values obtained ($58.6 \pm 3.4\%$) for % TBW from a study on Indian males (15) using the ethanol dilution technique. Urea and its derivatives bind preferentially to intracellular water, and therefore overestimate TBW (17). Deuterium is the tracer of choice for the determination of TBW, as it is accurate, safe, is easy to administer and can be used on all groups of people, including high risk populations, such as pregnant women and children (18).

In conclusion, this study demonstrates that accurate measurements of TBW by the deuterium dilution technique gives values for TBW that are comparable to the other methods as well as values in other populations. In addition, the deuterium dilution method can also be used to determine body composition.

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