

## Lean Body Mass in Indian Infants and Young Children and Variation by Sex

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**ABSTRACT** Aim of the study was to explore the lean body mass (LBM%) and fat mass (FM%) percent of weaning age children from among the urban poor in Kolkata, India. We used a cross sectional study design. A convenience sample of apparently healthy infants aged 6 month to 24 month were evaluated for LBM% derived by an anthropometry based and a Bioelectrical Impedance analysis (BIA) based equations, validated earlier on a sample of weaning age infants from this study population. Four hundred children (200 boys and 200 girls) from among the urban poor participated in this study. We measured their length to the nearest 0.1 cm and weight to the nearest 10 gm. Total body resistance was measured by a multifrequency BIA at 50 KHz. The calculated mean LBM% were 85.21 and 81.62 by anthropometry equation and, 82.72 and 82.56 by BIA equation in boys and girls respectively. LBM% values were considerably higher for both boys and girls compared to reference data on infants from USA. Based on weight for length, weight for age, length for age SD-scores stunting (21% boys, 11% girls), underweight (32.5% boys, 25.5% girls) and wasting (17% boys, 13.5% girls) were present using <-2 SD score as cut off for each. The LBM% was consistently higher in these infants aged 6 to 24 month compared to reference data on well infants in the West. Significantly higher proportion of boys had severe wasting than girls in weaning age infants in India.

### INTRODUCTION

A very rapid postnatal growth occurs in infancy and is accompanied by major changes in body composition. Knowledge of these changes in body composition in infants is useful in understanding the nutritional needs and functional outcome of nutritional management for healthy and sick infants (Koo 2000). Recent studies have generated new interest on the relationship between early nutrition and the future health of humans (Barker 2008). Both poor growth during early life and a large weight gain during infancy have been associated with disorders in adulthood. Repeated and accurate assessment of body composition during infancy enables us to determine the composition of the weight gained over time and provides key information for evaluating nutritional requirements, the efficacy of diet and medical interventions, and the influence of chronic diseases (Olhager et al. 2003).

Studies on how the state of nutrition interacts with the process of growth and the composition of incremental weight during early life in humans are important.

The two-compartment molecular level model in which body weight is divided into FM and LBM is the most widely used model in adults. The two widely used two-compartment models are based on the assumption that LBM has constant density and hydration. Although the LBM density and hydration are considered to be constant in healthy adults, they are not constant in infants and young children. Butte et al. (2000) reported a study designed to provide reference body composition estimates in infants and young children by using multi-component models. Butte et al. (2000) derived values for the hydration, density and potassium content of LBM in infants and young children. This pediatric modeling approach extends earlier pediatric models reported by Fomon et al. (1982).

Cadaver analysis being the gold standard for body composition analysis, no in vivo method can meet the highest standard of accuracy. Techniques for body composition such as underwater weighing are not feasible in infants and young children. Use of methods that involve any degree of radiation (e.g. DEXA radioactive tracers) is also not desirable for infants and small

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children. We have earlier used a stable isotope dilution technique as the reference standard to evaluate published equations for measuring total body water (TBW) based on anthropometry and on bioelectrical impedance analysis (BIA). Isotope dilution methods use a two component model to measure LBM and FM and are generally safe, reliable, accurate and feasible in infants and children. While they can be used in clinic and field setting, expense and expertise limits such use. Measurement of TBW with isotope dilution method has been used as a reference method in many of the classical studies of body composition (Fomon et al. 1982; Haschke et al. 1981; Forbes 1987; Fomon et al. 2002). To derive LBM from TBW one has to use age and sex specific hydration factor for LBM derived by multicomponent models (Butte et al. 2000; Fomon et al. 1982). While the hydration of LBM changes with age and maturation, use of age and sex specific hydration factors largely avoids errors associated with maturation (Cameron 2004). As stated earlier, Butte et al. (2000) have recently provided reference data for hydration factor for ages 15 days to 24 months by using an ambitious multicomponent model that provides age and sex specific reference hydration factors for LBM along with their confidence intervals. Their reference data now allows use of 2 component models using a stable isotope like  $D_2O$  in infants and young children with considerably more confidence.

Measuring FFM or LBM in infants and children is of both scientific and public health interest. Recent findings on the relationship between IUGR and diseases in adults such as hypertension, type II diabetes, and CHD generated renewed interest in studying the developmental indicators of infants and young children (Barker 1995, 1999, 2001; Eriksson et al. 2003). Further, it has been shown that more rapid postnatal growth (catch-up growth) in infants born with IUGR plays an important role on the risk of these adult diseases (Barker et al. 1993; Eriksson et al. 1999; Lucas 1991; Singhal and Lucas 2004). In the present study we have measured the LBM in 6 months to 2 years old urban infants and children from low socio-economic group using anthropometry and Bioelectrical impedance analysis (BIA) equations validated earlier (Sen et al. 2009; Sen et al. 2009 unpublished) for this population and investigated the pattern of percentage LBM (LBM%) in boys and girls. We have earlier evaluated

published equations based on anthropometry and BIA for children using  $D_2O$  dilution as reference method on infants and children in West Bengal (Sen et al. 2009; Sen et al. 2009 Unpublished). We have found that two equations, one based on anthropometry (i.e. of Morgenstern et al. 2002) and the other based on BIA (i.e. of Fjeld et al. 1990) are suitable for use in our study children.

In this paper we have explored the changes in LBM during the weaning period of infants and children from among the urban poor families. We have also compared the differences if any for LBM between boys and girls at such an early age of development.

## METHODOLOGY

**Subjects and Methods:** The study was conducted in apparently 400 healthy children (200 boys and 200 girls) from among the urban poor attending an immunization clinic of a large charitable government hospital in the city of Kolkata. The hospital service is provided free. The eligibility criteria for inclusion in the study were, age 6 to 24 months of either sex, absence of any reported illness during the preceding one month, absence of gross congenital anomalies and chronic diseases and parents willing to participate. The socio-economic and demographic features of the families are given in table 1. These children were from low socio-economic status and records on birth weight and gestation were not available for consideration. Written informed consent was obtained from the parents and the study was approved by the ethical review committee of the society for Applied Studies, Kolkata, India.

**Sample Size:** We calculated sample size to evaluate the difference in LBM% between boys and girls. Based on our earlier studies we wished

**Table 1: Socioeconomic status of the families**

Variables	Number (%)
House with cement floor, wall & roof	62 (15.2%)
Lives and cooks in one room	271 (68%)
family income per month(median)	Rs. 2000/-
Currently breast fed	358 (89.5%)
Mothers education	
Illiterate	75 (18.75%)
1-5 years of school	298 (74.5%)
6-10 years of school	27 (6.75%)
>10 years of school	0

to detect a difference of 2% in LBM% with a standard deviation of 5 between boys and girls with 95% confidence and 80% power. The estimated sample size of each group is 100. To detect a difference of 1.75% in TBW with the same assumptions the sample size is 131 in each group (The total would be 262). To detect a difference in LBM% of 1.5% with a standard deviation of 5 with the same assumptions we need a sample of 176 in each group. To allow for withdrawal of consent during the study we decided to recruit 200 boys and 200 girls for this study.

**Anthropometric Measurements:** All measurements were taken in the morning. The anthropometric measurements were made using recommended protocols (Lohman et al. 1988; Cameron 2004) and are briefly described.

**Length:** Recumbent length was measured with a wooden measuring board as described earlier (Shaikh et al. 2002). The board was made sufficiently broad to cover the shoulder blades. The reading was taken to the nearest 0.1 cm.

**Weight:** Weight was measured nude, using an electronic platform balance with a precision of 10 gm. The balance was checked regularly for accuracy using standard weights.

We used the anthropometry based best fitted equation by Morgenstern et al. (2002). They are:

$$\text{For Boys: TBW(kg)} = 0.0846 \times (L \times W)^{0.65} \dots (1)$$

$$\text{For Girls: TBW(kg)} = 0.0846 \times 0.95 \times (L \times W)^{0.65} \dots (2)$$

Where L= length in cm, W= weight in kg. These equations were derived from studies on the US subjects and aged 3 months to 13 years.

The LBM was calculated as TBW divided by an age- and sex-specific hydration factor for LBM (Butte et al. 2000).

**BIA Method:** BIA measures impedance of the body to a small electric current. The generic theoretical model treats the body as a single cylinder, with measurements made between electrodes placed manually on the wrist and ankle. Adjustment of bioelectrical data for height allows estimation of TBW. In our study, impedance was measured with a multi-frequency bioelectrical impedance analyzer (Xitron model 4000b; Xitron technologies Inc. San Diego, USA) using a single frequency of 50 kHz. Children with dry light clothes lay supine (Deurenberg et al. 1988; Deurenberg et al. 1889) with arms apart from the body and legs separated so that the thighs did not touch. After cleaning the skin contact area with alcohol, one pair of electrodes (foil

disposable 5 cm<sup>2</sup> ECG electrodes) was placed on the dorsal surfaces of the right hand at the distal metacarpal joints and between the distal prominence of the right radius and ulna. Another pair was placed at the distal metatarsal joints and between the lateral malleoli of the right foot. Resistance (R) value was recorded and used to calculate the total body water (TBW) using the following equation (Fjeld et al. 1990)

$$\text{TBW(kg)} = 0.76 + 0.18 \times (L^2/R) + 0.39 \times W \quad (3)$$

Where L= length in cm and W= weight in kg and R is resistance at 50 KHz.

The LBM was calculated as TBW divided by an age- and sex-specific hydration factor for LBM (Butte et al. 2000).

This equation was derived from studies on Peruvian children aged 3-30 months.

**Statistical Analysis:** Statistical programs Epi Info™ (CDC, Atlanta, USA, Epi 1994) and Stata™ version 7.0 (Stata Corporation, 4905 Lakeway Drive, College Station, TX 77845, USA) were used (Dibley 1987; Stata 1985). For anthropometric data, a software package based on the NCHS data base as provided with the Epi Info software was used. These anthropometric calculations were based on the growth reference curves developed by the NCHS and CDC using data from the Fels Research Institute and US Health Examination Surveys (WHO 1986). These growth curves are recommended by the World Health Organization (WHO) for international use. Data entry and editing were done using Epi Info Version-6 in a desktop computer. Statistical analysis was done using Stata Version-7. Means were compared using t-test and proportions were compared using chi-squared test. Regression models were used for adjusted analysis. Box-plots and trend lines were used to understand trends and associations.

## RESULTS

The age distribution was similar between boys and girls. However, the proportion of boys in the age group of 6-9 months tended to be higher (p=0.092). A larger proportion of boys had weight for height SD score <-2 (p=0.006) suggesting that the boys are more wasted (Table 2). A similar trend is noted for weight for age, the proportion with <-2SD score being higher in boys (p=0.12); similarly for height for age the proportion with <-2SD score showed a trend of being higher in boys (p=0.33).

LBM% are generally very high compared to reference data for infants in USA (Fig. 1). We have

**Table 2: Characteristics of subjects:**

Characters	Boys (n=200) Mean or number, SD	Girls (n=200) Mean or number, SD	p-value
Age(month)	12.64, 5.16	13.35, 5.09	0.167
6-9 month	77 (38.5%)	61(30.5%)	0.092*
>9-12 month	38 (19%)	42 (21%)	
>12-18 month	49 (24.5%)	59 (29.5%)	
>18-24 month	36 (18%)	38 (19%)	
Length (cm)	73.87, 5.71	73.37, 6.19	0.401
Weight (kg)	8.10, 1.41	7.82, 1.48	0.057
Resistance	757.61, 86.91	798.75, 76.64	<0.001
<sup>1</sup> WHZ	-1.10, 1.07	-0.97, 0.92	0.219
WHZ <-2	42 (21%)	22 (11%)	0.006*
WHZ >=-2 and <-1	70 (35%)	82 (41%)	
WHZ >=-1	88 (44%)	96 (48%)	
<sup>2</sup> WAZ	-1.47, 1.10	-1.34, 1.10	0.236
WAZ <-2	65 (32.5%)	51 (25.5%)	0.12*
WAZ >=-2 and <-1	73 (36.5%)	82 (41%)	
WAZ >=-1	62 (31%)	67 (33.5%)	
<sup>3</sup> HAZ	-0.98, 1.15	-0.83, 1.40	0.228
HAZ <-2	34 (17%)	27 (13.5%)	0.33*
HAZ >= -2 and <-1	60 (30%)	67 (33.5%)	
HAZ >= -1	106 (53%)	106 (53%)	

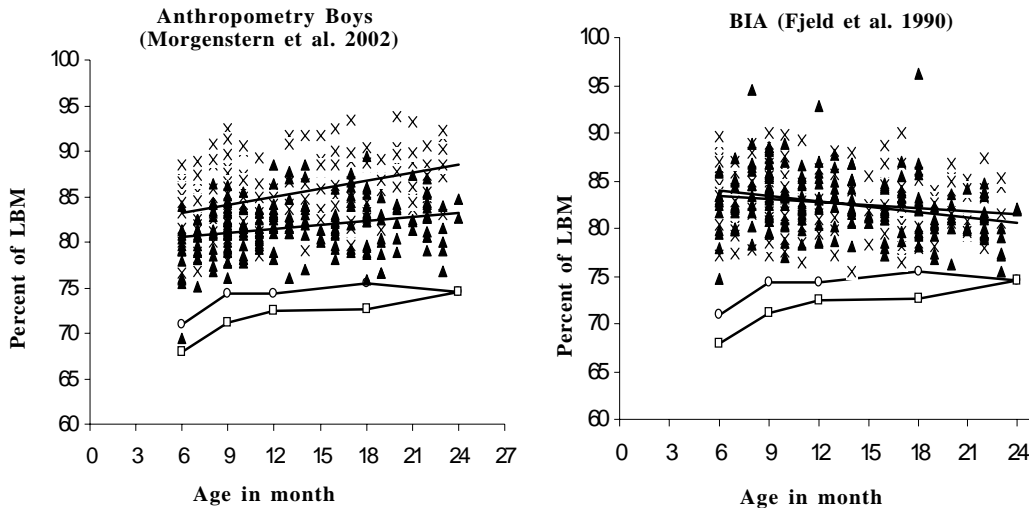
<sup>1</sup>WHZ, <sup>2</sup>WAZ, <sup>3</sup>HAZ are weight for height, weight for age and height for age standard deviation scores respectively compared to National Center for Health Statistics (NCHS) reference.

\* Chi-square test

compared the LBM% of the boys with the girls derived by the two equations based on anthropometry and BIA (Table 3). LBM% derived by the anthropometry equation is higher for boys ( $p<0.001$ ). But for the BIA equation, this difference

is very small and not significant. The trend lines in Figure 1 also illustrate this point.

Based on the anthropometry equation the lean body mass percent increased with increasing age in boys (Fig. 2a,  $p<0.001$ , ANOVA). However,



**Fig. 1.** Individual data points for LBM percent derived by the best fitted equations, (a) anthropometry based equation (Morgenstern et al. 2002) and (b) BIA based equation (Fjeld et al. 1990) are plotted against age in months. Age- and sex-specific reference values of LBM percent in healthy American infants derived by multicomponent models (Butte et al. 2000) are plotted for comparison (data points are connected). Boys (x), Girls (s), boys (○, Butte et al. 2000), girls (□, Butte et al. 2000). Trend lines for LBM percent in boys and girls are shown.

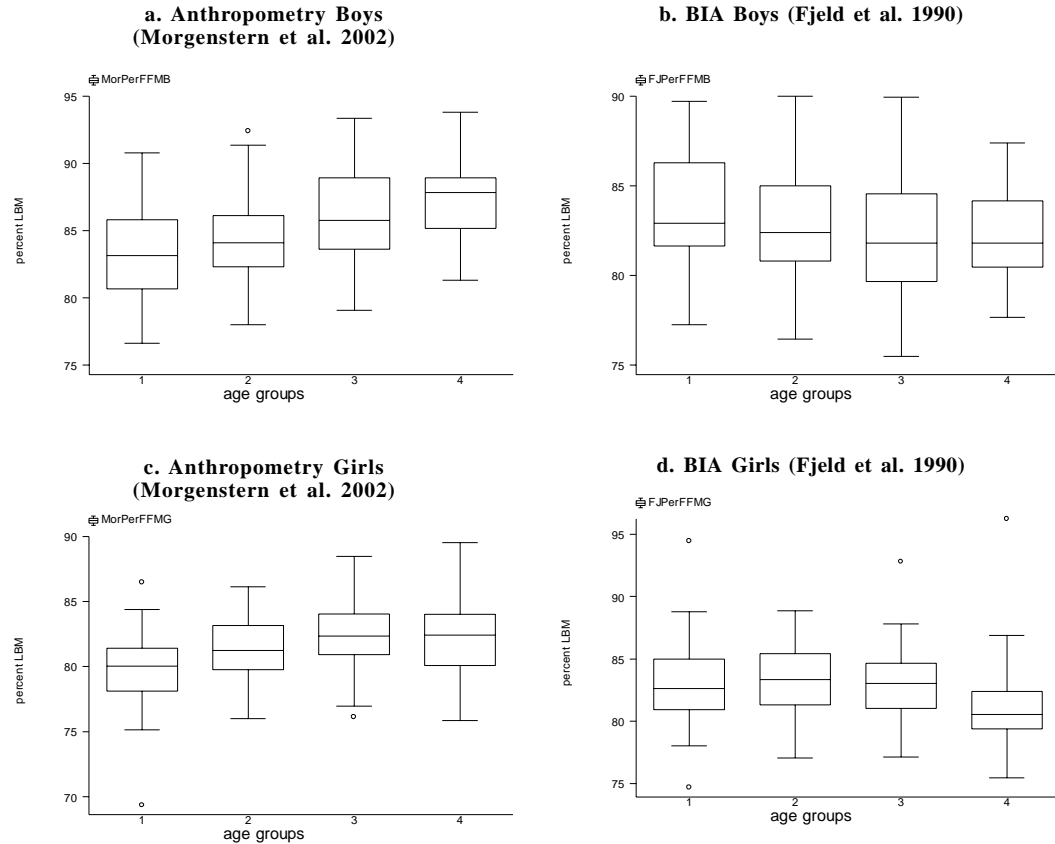
**Table 3: The mean and SD of LBM% of body weight derived by the two equations**

Methods	Boys (n=200) Mean or number, SD	Girls (n=200) Mean or number, SD	P-Value
Anthropometry (Morgenstern et al. 2002)	85.21, 3.53	81.62, 2.93	<0.001
BIA (Fjeld et al. 1990)	82.72, 3.18	82.56, 2.57	0.941

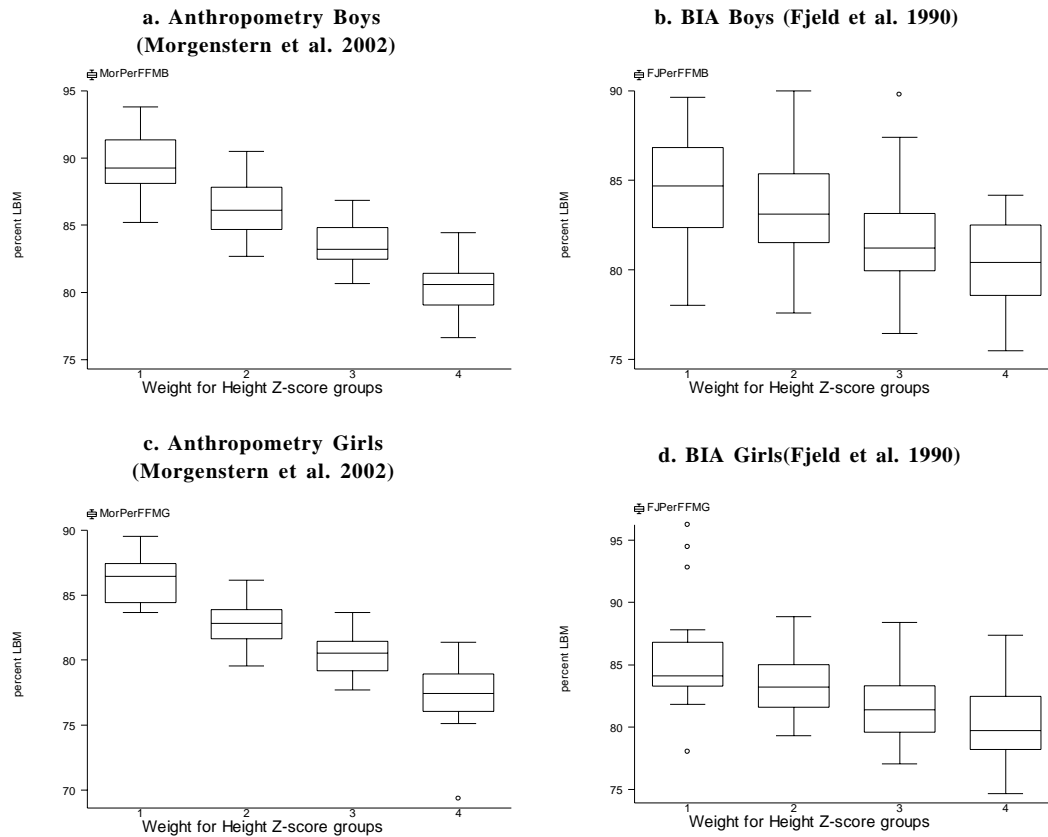
based on the BIA equation this trend was not seen (Fig. 2b,  $p=0.09$ , ANOVA). In girls also the LBM% consistently increased ( $p<0.001$ , ANOVA) when we used the anthropometry equation (Fig. 2c). But using BIA equation there is a significant nonlinear relationship (Fig. 2d,  $p=0.001$ , ANOVA). The LBM% increased upto 12 months, then

formed a plateau till 18 months. After 18 months of age LBM% decreased.

The conventional way to assess wasting or thinness is to compare weight for height with the reference data of the same age and sex. This is usually expressed as weight for height SD score or percentile. We compared the LBM% derived



**Fig. 2.** Box plots of LBM% derived by the anthropometry based (Morgenstern et al. 2002) and BIA based (Fjeld et al. 1990) equations by age groups in boys (2a, 2b) and girls (2c, 2d) are shown. The age groups are, 6 to 9 month =1(boys: 44, girls: 36), >9 to 12 month =2 (boys: 65, girls: 54), 12 to 18 month =3 (boys: 47, girls: 58), >18 to 24 month =4 (Boys: 44, girls: 52). The line in the middle of the box represents the median. The box extends from the 25<sup>th</sup> percentile to 75<sup>th</sup> percentile. The lines emerging from the box are upper or lower adjacent rules which extend  $\pm 1.5$  times the interquartile range.



**Fig. 3.** Box plots of LBM% derived by the anthropometry based (Morgenstern et al. 2002) and BIA based (Fjeld et al. 1990) equations by weight for height Z-score (WHZ) groups in boys (3a,3b) and girls (3c,3d) are shown. The WHZ groups are,  $WHZ < -2 = 1$  (boys: 42, girls: 22),  $WHZ \geq -2$  to  $< -1 = 2$  (boys: 70, girls: 82),  $WHZ \geq -1$  to  $< 0 = 3$  (boys: 55, girls: 69) and  $WHZ \geq 0 = 4$  (boys: 33, girls: 27). The line in the middle of the box represents the median. The box extends from the 25<sup>th</sup> percentile to 75<sup>th</sup> percentile. The lines emerging from the box are upper or lower adjacent rules which extend  $\pm 1.5$  times the interquartile range.

by anthropometry and BIA equations of boys and girls according to weight for height SD scores (Table 3). The 4 categories of increasing weight for height SD-score (WHZ) we used were  $WHZ < -2 = 1$ ,  $WHZ > -2$  to  $< -1 = 2$ ,  $WHZ > -1$  to  $< 0 = 3$  and  $WHZ > 0 = 4$  (Fig. 3,  $p < 0.001$ , ANOVA). The LBM% derived by both anthropometry and BIA equations consistently decreased and therefore FM% increased with increasing weight for height SD-score.

## DISCUSSION

The LBM% derived by validated equations based on anthropometry as well as on BIA is consistent with the conventional method of

assessing leanness such as weight for height standard deviation scores. Using either method lean body mass percent was inversely proportional to weight for height Z-score. Higher the LBM% lower is the FM% and more wasted the child is.

LBM% derived by both BIA based and anthropometry based equations are substantially higher in these children than the reference children from USA (Fig. 1). This reflects the presence and the magnitude of wasting in these infants and suggests inadequate nutrition in the weaning period. It is of interest to note that the vast majority of them are breastfed and mothers traditionally breastfeed them for longer periods. However, the energy density and protein content

of the traditional weaning food are known to be low and the degree of wasting may largely reflect this feeding practice. Mothers from low-middle socioeconomic strata breastfeed their children and prolonged breast feeding into the 2<sup>nd</sup> year of life is very common.

The changes in LBM% with increasing age produced inconsistent results by the two methods. Using the anthropometry based equation the LBM% increased with age both in boys and girls, albeit at a lower rate in girls. However, the LBM% derived by the BIA equation did not show this consistent trend.

Generic problem with equations based on length/height and weight is that they do not distinguish fat and lean masses. In most children with obesity increase in FM is also associated with increase in LBM albeit at a lesser rate. Because of this relationship the equations based on height and weight are able to predict the LBM with reasonable confidence. Body mass index (BMI, calculated as an index of relative weight) is often expressed as a standard deviation score to take into account age and sex in children. Although correlated with fat percent (largely because both LBM and FM increase in obesity), BMI cannot distinguish fat and lean masses, and there is a twofold range of variation in fatness for a given BMI value in individual children (Wells 2000).

The theoretical basis of bioelectrical impedance analysis indicates that it is well suited to assess TBW (Lancet 1992). The method is based on the phenomenon that only water (containing electrolytes) in the human body can conduct electricity. Fat is relatively devoid of water and restricts the flow of current through it. At any given current frequency the impedance of a cylindrical body system is a function of its length and cross sectional area. Nyboer (1972) developed the theoretical relation which is as follows:

$$V \propto L^2 / Z$$

Where V is volume of the body water in the subject, Z is impedance, and L is the length of the subject.

Impedance to the flow of current in body tissues is a function of resistance (R) and reactance (Xc). Cell membranes act as small capacitors and thus offer a reactive resistance (i.e. reactance) to the flow of current. Based on electrical theory, current at relatively higher frequencies passes through both extracellular and intracellular fluid and can provide an index of

TBW. Thus, BIA method can distinguish between non-fat body mass (LBM) and fat mass (FM) (Tang 1997). Of the two anthropometry based and three BIA based equations was evaluated by us the one by Morgenstern et al. (2002) based on Anthropometry and the other by Fjeld et al. (1990) based on BIA were found to give the best prediction of TBW compared to the reference method (Sen et al. 2009). Butte et al. (2000) have recently provided reference data on the body composition of children aged 0.5 to 24 months from the USA. As expected the LBM% is consistently higher in the study infants (coming from low socio-economic group families) compared to those in USA (Sen et al. 2009).

To derive LBM, one has to use age- and sex-specific hydration factors such as the ones derived by multicomponent models (Butte et al. 2000; Ziegler et al. 1976). While the hydration of LBM changes with age and maturation, use of age and sex specific hydration factor largely minimizes errors associated with maturation. For this age group the hydration factor for LBM ranged from 80.7 to 77.0 for boys and 80.7 to 78.0 for girls (Butte et al. 2000).

Based on skin-fold measurement, Yajnik et al. (2003) have shown that Indian babies are not only small at birth they also have less muscle mass and relatively more fat mass, the so called 'thin fat baby' syndrome (Yajnik et al. 2002, 2003). They further showed that thin fat babies grow up to become thin fat adults with thinner limbs and high waist-hip ratio; they appear to be foetally programmed and predisposed to diabetes. He also showed that the smallness and thinness of Indian babies is present at birth and an unusual thin-fat body composition is associated with the insulin resistance syndrome (Yajnik 2004). The conventional weight for height indices and BMI per-centile are likely to be inadequate to understand the growth and development of LBM. On the other hand BIA measurements relate best with TBW, a good proxy for LBM in a steady state condition.

These published equations should go a long way in fulfilling the need for in-depth studies to understand the early origin (i.e. fetal, neonatal and early childhood) of adult diseases of great public health importance such as, diabetes, hypertension, coronary heart disease.

## CONCLUSION

While the degree of wasting as indicated by

anthropometric indices increases with increasing age in weaning age infants among the urban poor in India, a more direct measure of lean body mass percent derived by BIA does not show this phenomenon. Prolonged and predominant breastfeeding may play a role in modifying the adverse effect of inadequate weaning food. The body composition changes in the weaning age infants in this population should be studied further by using methods relatively independent of anthropometry such as, BIA.

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