

# **CHAPTER II**

## **MATERIALS AND METHODS**

## **The population under study**

The northern part of the state of West Bengal, India is separated from the southern part by the Farakka Barrage. The area covers the Gigantic Himalaya and Sub-Himalyan region as well as the plains. It extends over an area of 21332 km<sup>2</sup> which is about 24.00% of the state and has a population of 14.72 million (14, 72, 14940) individuals, which is a little less than 1/5<sup>th</sup> of the state's population (Census 2001; IAMR, 2002). North Bengal is the home to a large number of heterogeneous tribal populations, caste Groups and religious communities. The region has a number of Mongoloid tribal populations such as the Lepcha, the Bhutia, the Meche, the Rabha, and the Santhal). Initially it was the British who were instrumental in bringing these individuals belonging to the Proto-Australoid tribal communities from the Chotanagpur plateau of Bihar to North Bengal mid-19<sup>th</sup> century to be employed as workers in the tea gardens. They are now collectively referred to as 'Tea-labourer' in North Bengal (Bhadra and Chakraborty, 1997). The most important scheduled caste populations are the Rajbanshi, the Bhumali, the Namsudra, the Bhuiya, the Kaibarta and the Turia. The present study is proposed to be undertaken among preschool children (aged 12 month to 60 months) and their mothers for the evaluation of nutritional status and disease-related morbidity factors. The ethical guidelines for human experiments study standard by the Helenski Declaration in 2000 should utilized (Touitou et al., 2004).

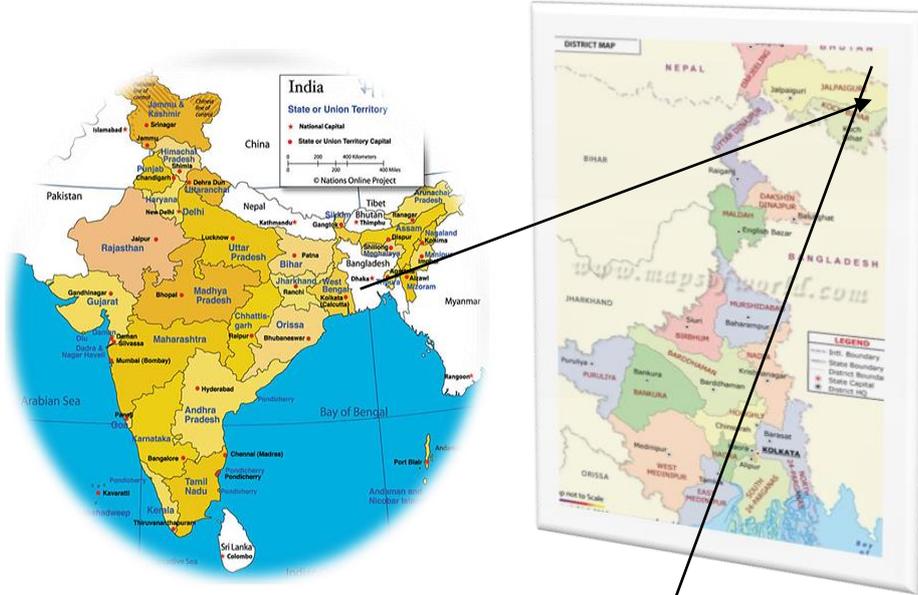
## **2.2. The study area**

Matigara (community development block) is an administrative division in Siliguri subdivision of Darjeeling district in the Indian state of West Bengal. Matigara Investigation Centre and Siliguri police stations serve this block. Headquarters of this block is at Kadamtala. There is one census town in this block:

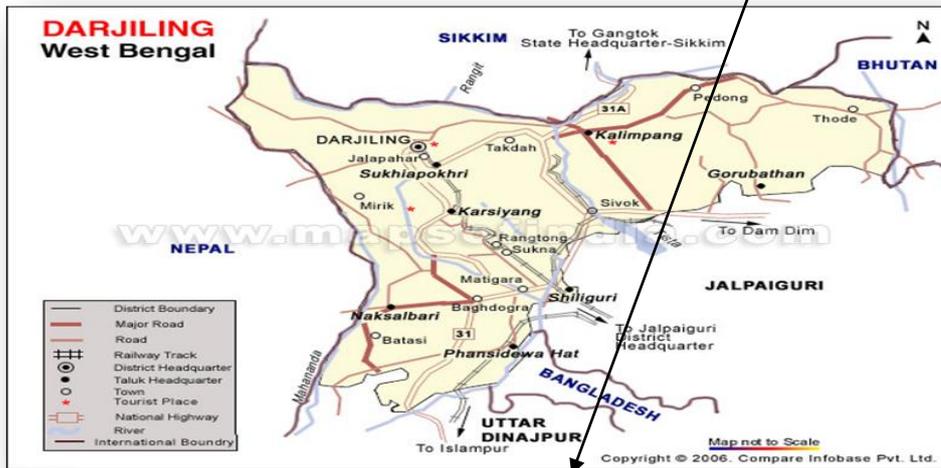
Bairatisal, Matigara is located at 26°43'0"N 88°23'0"E at an elevation of 127 m above sea level. Matigara community Development block has an area of 14.60 km<sup>2</sup>. Matigara block consists of one census town (Bairatisal) and rural areas with 5 gram panchyats, viz. Atharakhai, Matigara-I, Patharghata, Champasari and Matigara-II. The headquarters of this block is in Kadamtala. As per 2011 Census of India Matigara CD Block had a total population of 197,278 of which 135,583 were rural and 61,695 were urban. There were 101,023 males and 96,255 females. Scheduled Castes the population of were 70,527 and Scheduled Tribes were 26,484. As per 2011 census the total number of literates in Matigara CD Block was 129,006 out of which 72,352 were males and 56,654 were females. Matigara is one of the oldest weekly haat (market place) of Darjeeling District of Bengal. In early days elephant, horse, buffalo etc were sold here. People from different places even from remotest part of Nepal adjacent to Tibet boarder used come here for marketing. Douglas W. Fresh field in his classic "Round Kanchenjhangha" mentioned about this Hat. Pandit Sarat Chandra Das during his Tibet trip heard the story of this Haat from the villagers of Gunsha, Nepal in 1879 ([http://en.wikipedia.org/wiki/Matigara Bazar](http://en.wikipedia.org/wiki/Matigara_Bazar)). The haat serves people from all levels of society who come there to shop for their daily needs from in and around Matigara. In Matigara total ICDS project sanctioned are 315.

The present study was conducted among children residing in different villages, and also those frequenting the ICDS centers, under the Matigara Gram Panchayet (26°43'N 88°23'E), Police Station: Matigara, Sub-Division: Siliguri, District: Darjeeling, West Bengal. Children from Matigara, Chandmuni and New Chamta Tea Estates, Khapril More, Panhuman Basty and Nishchintapur are covered by these ICDS centers. The study area is situated at approximate distances of 8 km to 12 km from the sub-divisional town of Siliguri.

# WEST BENGAL



# INDIA



# MATIGARA

# MAP OF MATIGARA

### **22.3 Method of sampling and sample size**

Since the present study was a longitudinal study, the set of anthropometric measurements need to be recorded repeatedly over time. Children frequently the ICDS centers were the Case Group in the field setting was also needed Control Group. For this a Control Group in the field setting is also needed. The data was collected using a stratified random sampling method. Initially children belonged to the “Tea labourer Group” were identified. This was confirmed from the official records. Then children of the “Tea-Labourer” Group aged 12-60 months. Data was collected from the children and their mothers utilizing both ICDS and home visits. Initially 325 children and their mothers were approached to take part in the study. The study details were then explained to them. Subsequently, 6 months and their 10 children did not agree to take part in the study. Participation was purely voluntary.

Data was collected over 4 Phases extending over a period of 1 year. Phase I had 315 Case and 319 Control children. Phase II had 308 Case and 240 Control children, Phase III had 297 Case and 230 Control children and Phase IV had 248 and 201 Control children of Case Group and Control Group. The 248 Case and 201 Control Children of Phase IV were common in all the 4 Phases (purely longitudinal).

### **2.4 Procedures of data collection**

Standard procedures of Data collection were taken into consideration to obtain data from the field situation. The data collection procedure are briefly described below. The socioeconomic and demographic features of these preschool children and their mothers were collected by door to door surveys and the ICDS centers (for the Case Group) and door-to door surveys among those not visiting the ICDS centers (for the Control Group) using structured schedules.

### **Demographic, socio-economic and life style factors**

The data about the different socio-economic and demographic variables The socio-economic status of the subjects shall be assessed by utilizing the modified scale of Kuppuswami as proposed by Mishra and Singh (2003).

### **Demographic variables**

The explanatory variables that summarize the demographic behavior of a population are considered as demographic variables. In the present study both qualitative and quantitative variables were considered to assess their effects on nutritional status. The demographics variables examined were age, marital status, and dependent children family type. The household were also classified according to monthly family income, based on the classification provided by Chakraborty and Bharati (2010).

### **Socio-economic variables**

The variables which reflect the social and economic status of any population or Group are known as socio-economic variables. These variables are important to the assessment of the nutritional status. The socio-economic variables utilized using the modified scale of Kuppuswami proposed by Mishra and Singh (2003) and Kumar et al., (2007).The socio-economic scale has been described in Table A.

### **Life style related variables**

The life style related variables have added influence on the nutritional status of an individual or population. The variables recorded in the present study were source of drinking water, toilet facility and house type.

**Table 2.1: Kuppuaswami Soci-economic scale proposed by Mishra and Singh (2003) and Kumar et al., (2007)**

<b>(A) Education</b>	<b>Score</b>
1. Professional or Honors	7
2. Graduate or post graduate	6
3. Intermediate or post high school diploma	5
4.	
5. High school certificate	4
6. Middle school certificate	3
7. Primary school certificate	2
8. Illiterate	1
<b>(B) Occupation</b>	
1. Professional	10
2. Semi-professional	6
3. Clerical, shop owner and farmer	5
4. Skilled worker	4
5. Semi-skilled worker	3
6. Unskilled worker	2
7. Unemployed	1
<b>(C) Income per month (in Rs.) (as a modified by Kumar et al., 2007)</b>	
• 19,575 >	12
• 9,788-19,574	10
• 7,323-9,787	6
• 4,894-7,322	4
<b>Cont.: Income per month (in Rs.) (as a modified by Kumar et al., 2007)</b>	<b>Score</b>
• 2,936	3
• 980-2,935	2
• <979	1
<b>Socio-economic status</b>	<b>Total score</b>
Upper (I)	26-29
Upper-middle (II)	16-25
Lower-middle (III)	11-15
Upper lower (Lower)	10-5
Lower	<5

## **2.5 Nutritional assessment using anthropometric measurements**

Before obtaining the anthropometric measurements full explanation, procedure and objective of the present study were described to the each and every child and their mothers subsequently a verbal consent was taken from them prior to taking the measurements. All the individuals were not suffering from any diseases at the time of recording the measurements. They were free from any physical deformities and abnormalities.

### **Anthropometric measurement recorded**

Since the present study was a longitudinal study, the set of anthropometric measurements need to be recorded repeatedly over time so as to evaluate the effects of the nutritional supplementation given to the children (Case Group). For this a Control Group in the field setting is also needed. The measurements shall be taken three times at four monthly intervals over a period of one year. The anthropometric measurement was recorded following the standard techniques of Weiner and Lourie (1981), Singh and Bashin (1989) and Lee and Neiman (2005). Each measurement is briefly enumerated below:

#### **Height**

Height of the individuals was measured with the help of Anthropometer rod (GPM type, Galaxy Informatics, New Delhi) to the nearest 0.1 cm. They were made to stand on a horizontal platform (plastic board) with the Frankfurt horizontal plane. The horizontal arm of the anthropometry was brought down lightly to touch the vertex.

#### **Weight**

Body weight of the individuals was recorded using a portable weighing machine (Libra®, Edryl-India, Tiswadi, Goa). The individual were barefooted and wearing minimum clothing at the time of measurement. Body weight was recorded

to the nearest 0.5 kg. The weighing machine was checked regularly against a standard weight after weighing 15 individuals.

### **Mid Upper Arm Circumference (MUAC)**

Mid Upper Arm Circumference (MUAC) was measured on the left arm of each individual with the arm hanging relaxed. It was measured midway between tip of the acromion and the olecranon process. The measurement points were marked by a marker and the measurements taken with the help of a Gulick measurement was recorded in such a way that the tape was touching the skin but not compressing the tissue.

### **Triceps Skinfold (TSF)**

Triceps Skinfold (TSF) was taken on the triceps at a point marked at 1 cm above the midpoint between the tip of the acromion and the olecranon process. The elbow was placed at right angles to the middle of the upper arm and marked. The skinfold was pinched vertically and lifted off the muscle about 1 cm above the marked line. The measurement was then recorded.

### **Sub-Scapular Skin fold (SSF)**

Sub Scapular Skinfold (SSF) was measured under the angle of the left scapula. The skinfold was kept vertical or pointing slightly downwards and outwards. The skinfold was pinched vertically and the measurements was taken about 1 cm above the marked pointed.

### **Head Circumference**

Head circumference was measured by the standard anthropometric procedures (Hall et al., 2007) using a flexible, non-stretch plastic coated Gullick tape. Exerting light pressure, the tape was passed over the glabella to the area near the top of the occipital bone (opisthocranium) as to get the maximum circumference.

Care was taken to keep the tape flat against the head and parallel on both the sides. The measurement was recorded to the nearest 0.10 cm.

### **Technical Error Measurement (TEM)**

The technical Errors of Measurement  $\{TEM = \sqrt{(\sum D^2/2N)}$ , D=difference between the measurements. N number of individuals measured} is usually determined to check the validity and reliability of the anthropometric measurements (Ulizashok and Kerr, 1999; Goto and Mascie-Taylor, 2007). For the calculation of intra-observer TEM, height weight, MUAC, TSF, and SSF were recorded from 50 children and their mother (Mother: 25; children 25). The measurements were taken thrice on each individual by the investigator. The co-efficient of reliability (R), which ranges from 0 (not reliable) to 1 (complete reliable), was subsequently determined. The result of intra-observer TEM and the R value are shown in **Table.2**. Very high R value ( $R > 0.980$ ) were obtained for the intra-observer TEM analysis. All the values of R were appreciably higher than the accepted cut-off value of 0.95 as suggested by Ulizashok and Kerr (1999). Hence, the recorded anthropometric measurements were reliable and reproducible and the TEM were not incorporated for the further analysis.

### **Assessment of nutritional status from the measurements recorded**

The nutritional status of the mother and child was assessed using standard anthropometric indicators and body composition indicators. These are described as follows:

#### **Body Mass Index (BMI)**

BMI is considered to be a suitable indicator to estimate the intensity of under nutrition or CED and over-nutrition (overweight and obesity) of an individual or a population. BMI has been calculated using the following standard equation of WHO (1995):

$$\text{BMI (kg/m}^2\text{)} = \text{Weight (kg)}/\text{Height (m}^2\text{)}$$

Nutritional status has been assessed using the internationally accepted BMI cut-off points as suggested by WHO (WHO 1995) (Table 2.3). For the screening of under nutrition, different CED grades of BMI were used. Studies have indicated the BMI value of  $< 18.50 \text{ kg/m}^2$  as the cut-off points to determine under-nutrition among different Indian and non-Indian population (James et al., 1988; Gibson, 1990; Ferro-Luzzi et al., 1992; James et al., 1994; WHO, 1995; Ferro-Luzzi and James, 1996; James et al, 1999; Khongsdier 2001, 2005; Le and Nieman, 2005; Bose et al, 2006 a, b, c; Chakraborty and Bharti, 2010; Keino et al., 2014; Larty 2015; Mohammad and Hussain 2015; Mansur et al; 2015; Karimi et al., 2016; Rohde et al., 2017). The WHO Expert Committee (WHO. 1995) has further suggested the following classification of CED based on BMI among Mother and their children (**Table No.2.4**).

#### **Stunting (low height for age)**

Height-for-age is a measure of achieved linear growth that can be used as an index of past nutritional and health status. Low height-for-age is defined as shortness and reflects either a normal variation or a pathological process involving failure to reach linear growth potential. The outcome of the latter process is termed stunting or The ICDS programme uses its vast network of primary childcare centers and workers-known as anganwadi workers-to monitor children's growth by weighing at regular intervals. Weight is then earlier plotted for each child on Indian Academy of Pediatrics (IAP) growth charts, which were based on Harvard unisex growth standards. Recently IAP has adopted new WHO 2006 growth charts. Gaining of insufficient height relative to age. Stunting results from the extended periods of inadequate food intake, poor dietary quality, increased morbidity, or a combination of these factors. Stunting fails to distinguish between deficits associated with past

event and one associated with a long term, continuing process, yet this differentiation has major implications for intervention (WHO 1995). In earlier study reported that low stature-for-age is generally highest during the second or third year of life (WHO, 1986). The circumstances where low length-for-age occurred in the first year of life, it is said to reflect a continuous process of failing to grow or stunting, whereas in older children, it reflects having failed to grow or being stunted (WHO, 1995). Beaton in 1990 reported that in many populations appropriate treatment of children under two years of age generally results in improved stature (length/height-for-age), but for older children treatment will most probably have little effect on the child's height-for-age status. Nutritional stunting is a common problem of the pediatric population especially in developing countries. Although it is a resolvable problem, it continues to be an important health issue. Stunting can be diagnosed when a child's height falls more than two standard deviations below the mean height for age. Stunting may be caused by genetic, hormonal, pharmaceutical, psychosocial and nutritional factors (Hizli et al., 2007, Bose et al., 2007, Dewey and Cohen 2007; Bose et al., 2008; De Onis 2009; Prentice et al., 2013; Giuliani et al., 2016; Tariku et al., 2016, ).

### **Underweight (low weight for age)**

Weight-for-age reflects body mass relative to chronological age. Low weight-for-age is described as lightness and reflects a pathological process referred to as underweight, arising from gaining insufficient weight relative to age, or losing weight. Weight-for-age reflects both weight-for-height and height-for-age. It fails to distinguish tall, thin children from those who are short with adequate weight. Thus children with low weight-for-age may be genetically short, or their lower weight-for-age may result from stunting or nutritional growth failure, this condition is characterized by lower height-for-age but a weight appropriate to their short stature

(WHO, 1995). Poor growth and under nutrition are common in children (Saqladi et al., 2008; Hrusachka and Hadley 2016; Hurley et al., 2016). This weight-for-age index is widely used to assess under nutrition for its simplicity in measurement (Fentahun et al., 2016.). It is the most widely used index for assessment of under nutrition in clinical practice and the only one used by the Integrated Child Development Services (ICDS) programmer in India.

### **Wasting (low weight for height)**

Weight-for-height measures body weight relative to height. It helps to identify children suffering from current or acute under nutrition and is useful when exact ages are difficult to determine. Low weight-for-height in children is described as thinness and reflects a pathological process referred to as wasting. It arises from a failure to gain sufficient weight relative to height or from losing weight. High weight-for-height children are termed overweight and arise from gaining excess weight relative to height or from gaining insufficient height relative to weight (WHO, 1995). One of the characteristics of wasting is that it can develop very rapidly and under favorable conditions can be restored rapidly. In stunted children, weight may be appropriate for length or height whereas in wasting, weight is very low for stature as a result of marked deficits in both tissue and fat mass. A combination of weight-for-height and height-for-age should always be used for children in those low-income countries where the prevalence of stunting is generally much higher than that of wasting (Waterlow, 1972; Gorstein et al, 1994). But on the other Case such studies revealed that wasting was significantly associated with the households' poverty, poor access to health services, lack of mutual decision - making on the care or treatment of their sick child between biological parents, closer birth interval, and poor exclusive breastfeeding practice. Thus, an organized effort should be made at all levels to improve infant and young child feeding, health

services, child birth spacing behavior, and exclusive breastfeeding practice of the poor rural population particularly mothers to curb the problems of child under nutrition (Egata et al., 2013; Egata et al., 2014; Karimi et al., 2016; Derso et al., 2017). Fronza et al., (2016) reported that the prevalence of acute malnutrition (wasting) among 50 million under five children causing 8.0 % global child death annually.

**Table 2. 2. Table showed the result of Technical Error of Measurement of the Anthropometric measurements recorded in the present study**

Name of the measurement	Intra-observer	
	TEM	Coefficient of Reliability
Weight	0.100	0.998
Height	0.066	0.994
MUAC	0.066	0.997
HC	0.201	0.998
TSF	0.137	0.985
SSF	0.012	0.999

**Table 2. 3: Cut-off points for assessing nutritional status of adult individuals as Specified by WHO (1995)**

Category	BMI value (kg/m <sup>2</sup> )
CED Grade III	<16.00
CED Grade II	16.00-16.99
CED Grade I	<17.00-18.49
Normal	18.50-24.99
Overweight	≥ 25.00-29.99
Obese	≥ 30.00

**Table: 2. 4. Classification of CED based on BMI for adult individuals as specified by WHO (1995).**

Prevalence of CED	BMI value (kg/m <sup>2</sup> )
Low prevalence	Warning sign:5-9 % of population with BMI<18.50
Median Prevalence	Poor situation: 10-19% of population with BMI <18.50
High Prevalence	Serious situation: 20-39% of population with BMI <18.50
Very height prevalence	Critical situation:≥ 40% of population with BMI < 18.50

### **Mid Upper Arm Circumference (MUAC)**

Nutritional status was also been evaluated in the present study using the internationally accepted cut-off points of MUAC. The use of mid-upper arm circumference (MUAC) has improved the ability of front-line health workers to screen and assess for acute malnutrition among children by increasing the reach and enhancing the quality of Community-Based Management of Acute Malnutrition (CMAM) services (Collins et al. 2006, Brown et al. 2009). In 2009, the World Health Organization (WHO) and UNICEF published updated guidelines recommending a MUAC cutoff of <11.5 cm as one of three screening criteria for identifying and managing severe acute malnutrition in infants and children 6–60 months (WHO and UNICEF 2009). Although an earlier systematic review of the literature showed that children with MUAC measurements <11.0 cm had significantly elevated risk of mortality (Myatt et al. 2006), WHO and UNICEF

recommended a slightly higher cutoff to increase sensitivity of the measure, while maintaining high specificity. Largely due to the guidance from WHO and UNICEF on a standardized cutoff, MUAC has become a widely used and successful diagnostic tool for screening children and determining eligibility for services to manage acute malnutrition (Brown et al. 2009, Nyirandutiye et al. 2011). The individual values of MUAC found below 23 cm and 22 cm were characterized as under nutrition among the males and females respectively (James et al., 1994). The only study of adolescents we identified was conducted in India among nearly 2,000 adolescent girls (11–19 years) not attending school (Bulliyya et al. 2007), the vast majority (97 percent) of whom were anemic. Study revealed significantly lower mean hemoglobin levels in those with low MUAC (<22 cm) than those with MUAC  $\geq$ 22 cm. Among children MUAC < 115 mm and WHZ < -3 were used to define severe wasting as per the World Health Organization (WHO) classification (Bariend and Zamiki 1986; Goosence et al., 2012; Myatt 2006; Tadesse 2017).

### **Composite Index of Anthropometric Failure (CIAF)**

The prevalence of under nutrition was evaluated using the three commonly utilized conventional anthropometric indices of stunting, underweight and wasting (WHO 1995) and the CIAF (Svedberg 2000; Nandy et al. 2005). As the interpretation of the three conventional indices involves a comparison with an international reference population to determine under nutrition, the data from the National Centre of Health Statistics (WHO 1983) has been utilized as the reference population for the evaluation of under nutrition. The justification for the use of such a reference population is based on the empirical finding that well-nourished children in all populations follow similar growth patterns (Habicht et al. 1974).

However, several authorities have expressed some concerns about the suitability of the NCHS reference data as the basis of comparisons for assessing the nutritional status of the children from developing countries (Martorell and Habich 1986; de Onis et al. 1997). Recently, the WHO published child growth standards for attained weight and height to replace the previously recommended NCHS/WHO (WHO 1983) reference data. These new standards were based on breast-fed infants and appropriately fed children of different ethnic origins, raised in optimal conditions and measured in a standardized manner (de Onis et al. 2007). These reference data were proposed so as to identify the individual at risk, assess response to intervention and facilitate international comparisons (de Onis et al. 2001). However, the NCHS reference data (WHO 1983) has been found to be consistent with these currently advocated international growth references for children (WHO 1995; de Onis and Habicht 1996). Given these issues, the NCHS data (WHO 1983) has been used to assess the magnitude of under nutrition in the present study. The prevalence of under nutrition has been subsequently assessed utilizing Z-scores according to the classification of the WHO (1995). A child having values below 2 SD of the reference median in the indices of stunted, underweight and wasted was classified as undernourished (WHO 1995). The combination of Svedberg's (2000) model of six Groups (stunted only, underweight only, wasted only, wasting and underweight, stunted and underweight and lastly stunted, wasted and underweight and Nandy et al. (2005) (underweight only) have been utilized for assessing under nutrition using the CIAF. The CIAF classification is depicted in **Table 3.23**).

**Table 2.5. Classification of children with based on CIAF**

Group Name	Description	Wasting	Stunting	Underweight
A	No failure	No	No	No
B	Wasting only	Yes	No	Yes
C	Wasting and Underweight only	Yes	No	Yes
D	Wasting, Stunting & Underweight	No	Yes	Yes
E	Stunting & underweight	No	Yes	Yes
F	Stunting only	No	Yes	No
Y	Underweight only	No	No	No

*\*Classification following Nandy et al., 2005.*

### **Assessment of body composition among child**

Under nutrition among children was documented by calculating Z-score values using the available age and sex-specific WHO child growth reference values of WHO 2010. Z-scores were calculated using the WHO Anthro Plus calculator. The values of height-for-age Z-score (HAZ), weight-for-age Z-score (WAZ), weight-for-height Z-score (WHZ), and body mass index for Z-score (BMIZ) between “-2 to -3” and “<-3” were considered as moderately and severely undernourished, respectively. The BMI was calculated following the internationally accepted standard equation of WHO (2005)

$$\text{BMI} = \frac{\text{Weight (kg)}}{\text{Height}^2 \text{ (m)}}$$

$$\text{Height}^2 \text{ (m)}$$

The prevalence of under nutrition among mothers was assessed following the international BMI cut off points for CED as proposed by WHO 2005.

### **Assessment of Fat Mass (FM) and Fat Free mass (FFM)**

The Slaughter et al. (1988) were used to estimate PBF:

$$\text{Boys} = 1.21 (\text{TSF} + \text{SSF}) - 0.008 (\text{TSF} + \text{SSF})^2 - 1.7$$

$$\text{Girls} = 1.33 (\text{TSF} + \text{SSF}) - 0.013 (\text{TSF} + \text{SSF})^2 - 2.5.$$

And VanItallie et al. (1990) were utilized to assess the proportion of Fat mass (FM),

Fat-free mass (FFM), Fat mass index (FMI) and Fat-free mass index (FFMI): FM

$$(\text{kg}) = (\text{PBF}/100) \times \text{weight} (\text{kg})$$

$$\text{FFM} (\text{kg}) = \text{Weight} (\text{kg}) - \text{FM} (\text{kg})$$

$$\text{FMI} (\text{kg}/\text{m}^2) = \text{FM}/\text{Height}^2 (\text{m}^2)$$

$$\text{FFMI} (\text{kg}/\text{m}^2) = \text{FFM}/\text{Height}^2 (\text{m}^2).$$

The following the study of Fenton and Sauve in 2007 that Z-scores can be calculated from the LMS parameters by comparing the child's measure with the median size for that age, and dividing the result by the standard deviation.

Inclusion of the L parameter in the calculation:

$$\{z = (\text{measure}/M)^L - 1/ (L/S)\}$$

This formula takes any skew in the growth reference into account (Cole, 1989). The LMS parameters are expected to change smoothly with age since physiological changes are gradual and continuous, and differences from smoothness are usually due to sampling errors (Cole, 1989; Cole and Green, 1992). Smoothing of the L parameters can be justified based on these concepts (Cole and Green, 1992). Additionally, changes in L have very little impact on the skew of the final curves when the magnitude of the S values are small S, as they are in this Case for head circumference and length and also modified version. (Cole, 1989, Cole 1990).

At last the LMS Chart Maker software program (The Institute of Child Health, London) was used to obtain the smooth centile curves that fitted smooth centile curves to the reference data. The method summarizes percentiles at each age based on the power of age-specific Box-Cox power transformations used to

normalize data. The centile curves (3rd, 10th, 25th, 50th, 75th, 90th and 97th) were derived as reference data for further evaluation of body composition.

## **2.5 Statistical computation**

The data obtained in the present study was statistically analyzed using statistical constants and relevant statistical Package for Social Science (SPSS; version 17.0). A p-value of <0.05 and <0.01 was considered to be statistically significant.

Chi-square ( $X^2$ ) analysis was utilized to assess the prevalence of under nutrition among children in different category.

The Anthropometric variables recorded have been described using descriptive statistics (mean  $\pm$  standard deviation). Sex differences and age specific differences within sexes in the anthropometric variables were analyzed using one-way Anova analysis of variance (ANOVA). The Pearson correlation coefficient analysis was done to assess the dependency of the sex, age and BMI on the anthropometric variables.

$X^2$  analysis was also done to assess the sex differences in the anthropometric indices which are important in the assessment of under nutrition.  $X^2$  analysis was also performed to assess the sex difference in the non-parametric anthropometric features between the girls and boys individually. The Yates correction factor was taken into consideration if a cell/category possessed less than 5 individuals. This correction term adds to the accuracy of the  $X^2$  analysis when the numbers of classes are small,  $X^2$  analysis was also utilized to assess the sex differences in the nutritional indices related to fatness body composition, over-nutrition and regional adiposity measurement between the girls and boys measurement individuals. The differences in BMI with respect to the different

socioeconomic, demographic and life style related variables were also assessed using X2 analysis.

A multinomial logistic regression model was fitted to estimate the odds of being affected by stunting, underweight, wasted, low MUAC (Wasting) and also in low BMI for age (Thinness) in the categories of under nutrition among the children individually. The model with the calculation was fitted to estimate the odds of being affected by stunted, underweight and wasted, low MUAC for age and low BMI for age in the categories under nutrition among the children. The model with the calculation of corresponding adjusted odds (Ors) and 95% confidence interval (CIs) was used to examine possible difference between the individuals are fall in the three category of under nutrition. The predictor variables were socio-economic, demographic and life style related factors such as Age, Gender, Birth Order, Water facilities, Mother's age at child birth, Birth Interval, Mother's occupation, father's occupation, House Pattern, Media exposure, Sibling, family size, Toilet facilities, Electricity, Mother's Occupation, Father's Occupation, and Income. Case (Phase I, II, III and IV) and Control Group (Phase I, II, III and IV) of study analyzed one by one properly because each Phases are different to each other.

The multinomial regression model allows Controlling effects of these variables to create dependency in selective variables. Therefore, individuals affected by stunted, underweight, wasted (z-score based), low MUAC for age, and Low BMI for age (Thinness) were categorized dichotomously by using different cut-offs points for under nutrition. The individuals with any nutritional deficiency using a particular parameter such as stunted, wasted, underweight, low MUAC for age and Thinness categories were coded as 1. The individuals observed to be normal and also above the relevant nutritional cut-off points were coded as 0. These values were entered in the egression model as response variables and termed as dummy variables instead of

the actual values. Similarly, the predictor variables were coded separately and entered in the regression model as a set of dummy variables. The reference categories used for the multinomial regression model includes boys, 36-47 months, 1<sup>st</sup> birth order, 'no' water facility,  $\geq 19$  years mother's age at child birth,  $<24$  month birth interval, Illiterate mother's, Illiterate father's, pucca house in house type, regularly media exposures, 1-2 sibling category,  $\geq 4$  family size, 'yes' in Toilet facilities, 'yes' in electricity facility, those mother's were housewife, father's occupation 'others', income  $\geq 4001$ . The multinomial logistic regression model was tested separately for the boys and girls individuals. Same as using maximum likelihood estimation model was fitted to estimate the odds of being affected by Grades I, II, III. The predictor variables of Age, Gender, Birth Order, Water facilities, Mother's age at child birth, Birth Interval, Mother's education, father's education, House Pattern, Media exposure, Sibling, family size, Toilet facilities, Electricity, Mother's Occupation, Father's Occupation, and Income. A logistic regression using maximum likelihood estimation model was fitted to estimate the odds of being affected by wasting model allows the creation of categorical depended variables and the odds were obtained by comparing with reference category. Children who were both moderately ( $<-$  Moderate) and severely ( $<-$  Severe) undernourished were considered under the combined categories of under nutrition ( $<-$  Moderate). Therefore, children belonging to undernourished Group ( $<-$  Moderate) were coded as '0' and those with the higher or equal to  $-2$  SD (normal) were coded as '1'. These values were entered into the logistic regression model as response variables instead of the actual z-score values. Similarly, the predictor variables were coded separately and entered into the regression model as a set of dummy variables. A p-value  $<0.05$  was considered statistically significant.