CHAPTER 3

MATERIALS AND METHODS

The chapter discusses the site and the sample of the present study, the study design and indicators used for data collection, in accordance with the objectives.

3.1 Objectives

General Objectives

1. To conduct longitudinal studies on nutritional status of poor adolescent boys and girls in urban Vadodara

2. To undertake gender based analysis of the data to compare the nutritional status of adolescent boys and girls in different age groups from 10 to 18 years.

3. To identify environmental determinants of nutritional status of adolescent boys and girls: proximal and distal factors.

Specific Objectives

1. To study adolescent growth over one year in terms of height, weight, body mass index (BMI), mid upper arm circumference (MUAC) and skin fold thickness (SFT) in boys and girls.

2. To assess the prevalence of iron deficiency anaemia (IDA) among adolescent boys and girls through biochemical estimation of haemoglobin and to compare changes in Hb levels over 12 months.

3. To assess the prevalence and type of morbidities experienced by the adolescent boys and girls in different seasons: summer, monsoon and winter.
4. To determine gender differences if any, between the boys and girls with regard to the various nutritional health indicators mentioned above.

5. In the adolescent boys and girls included in the study, to assess environmental determinants which influence their nutritional status specifically various proximal and distal determinants given below:

* Quality and quantity of food consumed and intra-household food distribution in three seasons: summer, monsoon and winter.
* Perceptions regarding nutrition, health, gender based roles and responsibilities of adolescent boys and girls.
* Socio-economic factors such as education and occupation of parents, type of family, family size, religion and sources of health information.

3.2 Selection of Site

The study was conducted in one urban slum and two government aided schools catering to low socio-economic group. The slum was purposively selected and was typical of the most slums in Vadodara city in terms of basic civic amenities, ethnic groups (mainly Hindu families with fewer Muslims or other groups) and health care services. Earlier studies conducted on urban poor adolescent boys and girls in this department, had shown that most of the older adolescent boys (above 15 years) in slums were working and not easily available during data collection. As for the older adolescent girls (16 years and above), many were married and those not married were studying in nearby school. Therefore, to have a sufficient sample of 10 – 18 years old boys and girls (including the older adolescents) for the present study especially to study longitudinal growth, the government aided schools located near the selected slum were also included in this study. Visits to the schools and meetings with the school principals revealed that many of the slum children were studying in these two schools and socio-economic status of the school going adolescents was similar to the adolescents living in the slum. Thus it was possible to enroll adolescents of the school and near by slum in this study.
3.3 The Sample of the Study

All the children in the age group of 10 - 18 years from the selected schools and the slum were enrolled for the study (Fig 3.01). Written permission from the parents of the boys and girls enrolled was obtained (Appendix I). Socio-economic status and anthropometric surveys covered all the children who were enrolled and available at the time of these surveys. An assessment on prevalence of anaemia, morbidity profile, quality and quantity of food intake and perceptions of the adolescents about their own nutritional status, knowledge about health and nutrition issues and their role and responsibilities at the household level was carried out on a random sub-sample stratified by age and sex (explained later).

Only those subjects who were present throughout the study period and on whom longitudinal data (initial and after 12 months) could be obtained, were included in data analysis. Thus, the study sample comprised of 330 boys and 362 girls in the age group of 10 – 18 years. The number of adolescent boys and girls studied for the different age groups is presented in Table 3.01.

Table 3.01 : Distribution of 10 – 18 Years Old Boys and Girls by Age and Sex

<table>
<thead>
<tr>
<th>Age (Years)</th>
<th>Boys</th>
<th>Girls</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>10 - 12</td>
<td>98</td>
<td>98</td>
<td>196</td>
</tr>
<tr>
<td>13 - 15</td>
<td>142</td>
<td>173</td>
<td>315</td>
</tr>
<tr>
<td>16 - 18</td>
<td>90</td>
<td>91</td>
<td>181</td>
</tr>
<tr>
<td>10 - 18</td>
<td>330</td>
<td>362</td>
<td>692</td>
</tr>
</tbody>
</table>

3.4 Study Design

The study design and phases are depicted in Fig. 3.01. This was a longitudinal study carried out over a period of two years.

As the study design illustrates, the longitudinal data were collected to assess firstly growth in terms of anthropometry over a one year period and secondly, changes in Hb levels and prevalence of anaemia during this period. Further, longitudinal data were also collected by season, to assess variations in food and nutrient intake, intra-
**Fig 3.01: Study Design**

10 - 18 years old Adolescents

Boys (B) (N = 330) + Girls (G) (N = 362)

**Initial Assessment**

- Anthropometry B+G: 330+362
- Hb Estimation 90 + 96
- Socio-Economic Status: 330+362
- Perceptions of 15-18 years old adolescents: 35 + 42

**Data Collection by Season**

- **Seasons**: Summer, Monsoon, Winter
- **Indicators**
  - Morbidity Profile
  - Diet Surveys
    - Food Frequency
    - 24 hour recall covering intake of all family members
    - Intra-household food distribution
house hold food distribution and morbidity experienced during the summer, monsoon and winter seasons (Fig 3.01). In the beginning of the study, socio-economic status indicators and perceptions of the older adolescents (15 – 18 years) regarding roles and responsibilities were assessed. Table 3.02 and Table 3.03 give indicators, sample and methods used for data collection.

Table 3.02: Phases of Data Collection and Sample Sizes for Various Indicators

<table>
<thead>
<tr>
<th></th>
<th>Initial</th>
<th>At 12 months</th>
<th>By Seasons</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Summer</td>
<td>Monsoon</td>
</tr>
<tr>
<td>1. Socio-economic Status</td>
<td>B 330</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>G 362</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>2. Perceptions</td>
<td>B 35</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>G 42</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3. Anthropometry (Height, Weight)</td>
<td>B 330</td>
<td>330</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>G 362</td>
<td>362</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>4. Hb estimation*</td>
<td>B 90</td>
<td>-</td>
<td>118</td>
<td>118</td>
</tr>
<tr>
<td></td>
<td>G 96</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5. Morbidity*</td>
<td>B -</td>
<td>-</td>
<td>112</td>
<td>118</td>
</tr>
<tr>
<td></td>
<td>G -</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>6. Food frequency*</td>
<td>B -</td>
<td>-</td>
<td>94</td>
<td>98</td>
</tr>
<tr>
<td></td>
<td>G -</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>7. 24 Diet recall*</td>
<td>B -</td>
<td>-</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td></td>
<td>G -</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Random sub-sample B = Boys G = Girls

3.5 Description of the Parameters Used for Data Collection

3.5.1 Anthropometry

3.5.1.1 Height for Age

**Principle**: Height is a linear measurement made up of the four components, legs, pelvis, spine and skull (Gibson1989).

**Technique**: To measure height, a fibre glass tape was affixed vertically on a smooth wall in
the school and a community health centre in the slum, perpendicular to the ground, taking care to see that the floor area was smooth and not rough. The subject was asked to remove the shoes, stand with his/her centre of the back touching the tape, feet together and parallel, and with heels, buttocks, shoulders and back of the head touching the wall. The head was held comfortably erect, arms loosely hanging by the sides. A smooth thin ruler was held on top of head in the centre, crushing the hair at a right angle to the scale and height read off from the lower edge of the ruler to the nearest 0.5 cm. Each reading was taken twice to ensure the correctness of the measurement (Gibson 1989).

Table 3.03: Indicators of Nutritional Status Assessment among Adolescent Boys and Girls and Determinants of Nutritional Status

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Methods</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Socio-economic status</td>
<td>Semi structured pre-tested questionnaire</td>
<td>Bernard (1991)</td>
</tr>
<tr>
<td>• Perceptions regarding their own health</td>
<td>Semi structured pre-tested questionnaire</td>
<td></td>
</tr>
<tr>
<td>• Height, weight, mid upper arm circumference, skin fold thickness</td>
<td>Standard anthropometric methods</td>
<td>Gibson (1989)</td>
</tr>
<tr>
<td>• Prevalence of Anaemia</td>
<td>Estimation of haemoglobin by cyanmethaemoglobin method</td>
<td>Oser (1979)</td>
</tr>
<tr>
<td>• Prevalence of Morbidity by season</td>
<td>Semi structured pre-tested Questionnaire</td>
<td></td>
</tr>
<tr>
<td>• Quality and quantity of food consumed by boys and girls, intra-household food distribution by season</td>
<td>24 hour recall method and semi structured pre-tested questionnaire</td>
<td>Gibson (1989)</td>
</tr>
</tbody>
</table>

3.5.1.2 Weight - for - Age

Principle: Body weight is a measurement of muscle, fat, bone and internal organs. Weight deficits appear to be the best indicator of the prevalence of protein energy
malnutrition in the study population. Weight measurements when taken at a period of regular interval indicate a picture of growth in adolescents. Comparison of weight for age values with the accepted standards at corresponding ages reveals the degree of malnutrition in the adolescent boys and girls. Weight for age gives a picture of current nutritional status of the subjects.

**Technique**: An adult weighing balance (also known as bathroom scale) was used as it is portable and convenient to use in the field. The accuracy of the weighing machine was checked against 5 kg dead weight everyday morning before taking to the field for the measurements. The scale was adjusted to zero before each measurement. The subject was asked to stand on the platform of the scale without touching anything, with his/her head straight and eye looking in front. The subjects removed their socks and foot wears and wore minimum clothes viz. their school uniform or dress. The adolescents were asked to remove their watch/(bangle), belts and keys and wallets from the pockets before weight measurements were taken. Two readings were taken for accuracy and the weight was recorded to the nearest 0.25 kg in a register. The weight measurements were taken twice during the study period, first at the beginning of the study and second after one year of the study.

**3.5.1.3 Body Mass Index**

Body mass index (BMI) is considered an appropriate indicator of nutritional status in adolescence. By controlling height, BMI is considered an indicator of weight, that is, independent of height. BMI is expressed as the ratio of weight (kg) and height in meter square (weight (kg)/height(m)^2).

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

**Principle**: Muscle and fat constitute the soft tissues that vary with deficiency of protein and calories. Measurement of the mid upper arm circumference (MUAC) is a useful, practical method for assessing muscle mass, as this region is easily accessible and measurement requires only a flexible fibre glass tape.
Technique: The subject was asked to flex his/her arm at the elbow such that the lower arm was at right angle to the upper arm. The length between the acromion process of the scapula and olecranon process of the ulna was measured with a flexible fibre glass tape and the site of the measurement, exactly midway down the upper arm, was marked with a pen. The adolescent boy/girl was asked to hang his/her arm relaxed by the side and the tape passed gently, but firmly, around the arm at the selected mid-point, taking care to avoid compression of the soft tissues of the arm. The arm circumference was then measured twice to ensure accuracy to the nearest 0.1 cm.

3.5.1.5 Skin Fold Thickness (SFT)

Principle: Measurement of skin fold (or fat fold) at the triceps is one of the methods for assessment of the amount of subcutaneous fat, which gives an indication of the calorie reserves in the body.

Technique: The site selected for measuring MUAC was used, midway down the length of the left upper arm. With the arm hanging relaxed at the side and the skin fold parallel to the long axis was picked up between the thumb and forefinger, well away from the underlying muscle, and the skin fold measured to the nearest 0.1 mm by the means of the Harpenden calipers. The dial of the calipers was checked each day and reset to zero if necessary. Each measurement was taken twice to ensure accuracy.

3.6 Haemoglobin Estimation

Haemoglobin level of blood is used as an index for detection of anaemia. It was estimated by using a modified version of the Cyanmethaemoglobin method as given by Oser (1979).

Principle: Haemoglobin present in the blood sample reacts with ferricyanide to form methaemoglobin, which is converted to Cyanmethaemoglobin by cyanide. This complex is read colorimetrically at 540 nm.
Technique: The method was standardized as per the accepted procedures. A sterile lancet was used to prick the disinfected finger tips. The first drop of blood was wiped off using filter paper. Then 0.02 ml of blood was pipetted taking care to avoid the formation of air bubbles. The tip of pipette was wiped free of blood with a piece of filter paper. The collected blood was then transferred in to test tube containing five ml of Drabkin's reagent. The contents were mixed by twirling the test tube between the palms. The readings were taken on the same day using colorimeter at the wave length of 540 nm. Before initiating reading procedure, the instrument was set at zero using five ml of Drabkin's reagent as the blank. The readings were taken for each sample and haemoglobin as gram percent was calculated using the factor of 36 derived from the process of standardization (Appendix II).

3.7 Morbidity Status

It has been mentioned earlier that data on prevalence of morbidity among adolescent boys and girls is scarce. In the present study, an attempt was made to elicit data on prevalence, type and duration of morbidity and treatment seeking behaviour of the study subjects. The morbidity history was recorded for the 15 days prior to the survey. A semi structured pre-tested questionnaire was used (Appendix III). The morbidity profile was collected for the three seasons namely; monsoon, winter and summer.

3.8 Food and Nutrient Intake by Season

The food and nutrient intake of the subjects were assessed through food frequency and 24 hour recall method which are described below.

3.8.1 Food Frequency

Principle: This method can provide information where evidence is sought regarding a subject's diet in general rather than the diet over a specified time. The usual dietary intake of the subjects in terms of frequency with which various food items are consumed is recorded.
Technique: The subjects were asked to recall the frequency of consumption of protective foods by food groups on daily, weekly, fortnightly and monthly basis (Appendix IV). The protective foods whose frequency of intake was sought were pulses, legumes, GLVs, other vegetables, roots and tubers, fruits, milk and milk products and non-vegetarian foods.

3.8.2 24 Hour Recall Dietary Survey

Diet surveys provide information on nutrient adequacy or inadequacy of the subjects under the study. Adolescence being a period of rapid growth, there is a high demand for nutrients (Narsingrao 1985). This makes it imperative to measure nutritional adequacy along with their growth pattern during this period.

The 24 hour recall method was employed to collect data on quality and quantity of foods consumed by the adolescent boys and girls as well as intra-household food distribution. 24 hour recall data were collected for each subject in the three seasons during the study period.

Principle: This method involves the process of recalling food consumed by the subject on the day before the survey. The raw ingredients for each of the recipe recalled by the respondents are measured in volumetric cups. The volume of cooked food for the whole family and the cooked food consumed by the subjects are also recorded. Based on the volume of cooked food and amount of raw ingredients used, the individual intake of the raw food items is calculated.

Technique: The subjects selected for the study were informed about the purpose of the study. Their home addresses were noted down and home visits were made to collect information about the kinds of the foods and beverages consumed by the subjects and their family members on a previous day.

The respondents, that is, the adolescent and the mother were asked to report about the type and quantity of foods cooked for the whole family and amounts consumed by the each family member on the previous day of the survey. The volume of the total cooked food, the total amount of the ingredients used for cooking and amount...
of cooked food eaten by each member of the family was recorded using volumetric cups. This data was used to ascertain the amount of raw food items consumed by each of the family member by using the accepted procedures. The nutrient value of each raw food item was calculated using the food composition tables (Gopalan et al 1989). The nutrient intake was compared with recommended daily allowance (RDA) for Indians (Gopalan et al 1989).

The interview was conducted with specific probes. Probing was done to collect all the necessary details such as how many family members partook food on the previous day, whether guests were present or any of the family member had food outside the home (Appendix V).

**Intra-Household Food Distribution**

Intra-household food distribution (IHFD) among the family members was calculated for six nutrients namely, energy, protein, calcium, iron, vitamin A and C. To calculate IHFD, the following procedure was carried out.

1. **Percent RDA of each nutrient by all the family members**

   The percent RDA of each of the family members for a given nutrient was calculated, for example percent RDA for calorie of a family with five members. Mean % RDA values were then compared among various family members to ascertain the intake of adolescents vis-à-vis the other members.

2. **Scoring system using % RDA intake**

   To further look at the relative intake of various nutrients by each family members from the family pool of available food (that is, share from the family pot), a scoring system was developed.

   2.1.1 The percent RDA intake of all the family members was added to obtain family pool for a given nutrient. This value was divided by the family size to derive a factor X which represented the per capita availability of a given nutrient.
2.1.2 A score for each member was obtained by dividing the percent RDA intake by that member for a given nutrient by the factor \( X \). If the member scores more than 1, it means that the member has consumed more than what is available for him/her from the family pot. If it is less than 1 then he/she has received less than his/her due share from the family pot to meet his/her needs.

This is demonstrated below by an example:

<table>
<thead>
<tr>
<th>Family members (n=5)</th>
<th>% RDA (Calories)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult male</td>
<td>90</td>
</tr>
<tr>
<td>Adult female</td>
<td>85</td>
</tr>
<tr>
<td>Adolescent male</td>
<td>75</td>
</tr>
<tr>
<td>Adolescent female</td>
<td>70</td>
</tr>
<tr>
<td>Male child</td>
<td>72</td>
</tr>
</tbody>
</table>

\[
\text{Total} = \frac{392}{5} = 78.4
\]

\[
\text{Intra – Household Distribution Score}
\]

<table>
<thead>
<tr>
<th></th>
<th>( \frac{% \text{ RDA}}{78.4} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult male</td>
<td>( \frac{90}{78.4} = 1.15 )</td>
</tr>
<tr>
<td>Adult female</td>
<td>( \frac{85}{78.4} = 1.08 )</td>
</tr>
<tr>
<td>Adolescent male</td>
<td>( \frac{75}{78.4} = 0.96 )</td>
</tr>
<tr>
<td>Adolescent female</td>
<td>( \frac{70}{78.4} = 0.89 )</td>
</tr>
<tr>
<td>Male child</td>
<td>( \frac{72}{78.4} = 0.92 )</td>
</tr>
</tbody>
</table>

After calculating each member’s family share, independent “t” test was carried out on the mean scores to see whether the mean share of adolescent male and female was significantly different from the mean share of rest of the family.

3.8 Semi Structured Interviews

The interview tool was used to elicit information on socio-economic profile and perceptions of the adolescents on nutrition and health issues.

**Principle:** This interview is based on the use of structured questions that need to be covered in some order. There are also some open ended questions. The
questions should follow a pre-determined pattern but their substantive focus shifts according to the responses of the informants (Bernard 1991).

The technique to collect data pertaining to socio-economic profile and perceptions of the adolescents is described below.

3.9.1 Socio–Economic Profile

The semi structured questionnaire to assess the socio–economic status of the study subjects was prepared and pre-tested. The questionnaire was distributed in the classroom and the adolescent boys and girls were asked to fill it in the presence of the investigator. The filled in questionnaires were checked on the same day evening in order to get back to the subjects the next day, in the event of any discrepancy or incompleteness encountered. In the selected slum, the questionnaire was filled in by the researcher. The questions included birth date, caste, family size, age, sex, education, occupation and income of the family members, type of house they lived in. Availability of toilet facility by its type, water and drainage facility, type of fuel used, material goods such as TV, radio, fan, mixer, type of vehicles was also asked. The study subjects were also asked about their perceptions of the ventilation facility in their houses (Appendix VI).

3.9.2 Perceptions of Boys and Girls (15 – 18 years)

In this present study, perceptions of 15 – 18 years old boys and girls were assessed about their own

- nutritional status
- adolescent growth
- knowledge about balanced diet
- gender roles and responsibilities and its effect on their own health

The questionnaire was designed and developed in English, which was then translated in to Gujarati to enable the study subjects to understand and fill in by themselves. After translating into Gujarati, it was pre–tested (Appendix VII). The boys and girls aged 15 years and above and were willing to participate, were included for the survey. The questionnaire was filled in by the study subjects in the
presence of the investigator. The filled in questionnaire was checked on the same day and the subjects were contacted the next day for clarifications whenever required.

During the course of the study and interactions with the adolescents as well as in the filled in statements, several verbatim statements were recorded reflecting the adolescent views in their own words.

3.10 Analysis of Data

All data were analyzed keeping in view the study objectives. The quantitative data including diet survey were entered in the database package - FoxPro 2.5. For the analysis, FoxPro (2.5) and Epi Info 6.0 – an epidemiological package developed by WHO were used. The nutrient intake, and intra-household food distribution data was analyzed using FoxPro programmes. The details of the data analysis are given below.

3.10.1 Socio – Economic Status, Morbidity, Food Frequency and Perceptions

Percentages responses of the study subjects were calculated for the different variables in Fox Pro 2.5.

3.10.2 Anthropometry, Food and Nutrient intake

Statistical tests such as mean, median, standard errors and 't' test for comparison between the boys and girls for anthropometric measurements were performed using Epi Info 6.0 package. The percent standards for W-A, H-A and W-H were calculated using NCHS standards. The reference standards for MUAC and SFT reported by Vijayaraghavan et al 1974 were applied to calculate percent standards. The BMI percent standards were arrived by using the reference standards reported by Must et al 1991. The percent standard value for each subject and the percentages for prevalence of under-nutrition were determined in Fox Pro 2.5 package. As for food and nutrient intake, the % RDA was calculated by the programmes developed in the Fox Pro 2.5 using the RDA given by Gopalan et al (1989).
Chi square test was conducted to ascertain association between proximal and distal factors with nutritional status and gender differences viz., proportional difference in prevalence of under-nutrition between the boys and the girls, correlation between anaemia and morbidity, association between socio-economic variables and nutritional status. Independent ‘t’ test was applied to see the difference in nutritional status of adolescent boys and girls, relationship between nutrient intake and nutritional status, gender differences in mean values of anthropometric measurements and mean increments of Hb values.