Chapter - III

Materials and Methods
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MATERIALS AND METHODS

3.1 Location

The actual place of field investigation was Orchha block of Bastar district in Chhattisgarh state (Fig.3.1). The area is popularly known as Abujhmar and a prohibited place for visitors. Roughly elliptical in shape with longer axis in the north west to south east direction, Abujhmar tract is situated between latitudes 19°.0' and 20°.0' N and longitudes 80°.39' and 81°.39' E. It is bounded on the north by the plains of Antagarh and Narayanpur on the east by the country jharia Murias and by the Marhin (Gudra) river, on the south by the plain areas on the banks of Indrawati and on the west and north west by Kotri river and its tributary the Walter. West wards the tract extends beyond the borders of Bastar district into Chandrapur (Garhchiroli) district of Maharashtra. Area of this unknown tract is between about 1500 to 1600 square miles (Tiwari 1984).

3.2 General Scenario of Study Area

Abujhmarias inhabit Abujhmar high lands (unknown) in district Bastar of Chhattisgarh. The Abujhmar translates into English as “The unknown plateau”. The whole area covers about 4000 square kilometers. Its high ridges and deep valleys are cut into by the icy scalpels of many streams. These hills do not conform to any regular feature but there are hills and valleys in all direction. The only clues that help to a conception of its features are the ‘watersheds’. About 2800 square kilometers of it without any kind of road at all, and the very steep thickly forested slopes have to be negotiated by means of improbable trials. Such roads as do exist on the northern and eastern perimeters are inaccessible in the rains with steep gradients that are susceptible to landslides. It is not a friendly place, not even to the Marias who is its natural inhabitants. The Abujhmarias are a simple, shy and retiring people who suffer from geographical isolation of extreme degree. Abujhmar is spread over three tehsils.
Fig. 3.1: Map showing the location of study area (Orchha/Abujhmar) in Bastar district of Chhattisgarh state, India.
namely Narainpur, Bijapur and Dantewara. Every year there is an annual bazaar-cum religious assemblage known as marhi or madai at Narainpur some time in February and this marhi is attended by almost all villagers. General picture of the area with regard to the status on various aspects of Abujhmar scenario is briefly presented as follows:

3.2.1 Flora and fauna

The flora of Abujhmar is fabulously rich. Sal forests occur in the north up to Irkbbattai and in the south eastern corner. Mixed forests are common in the middle part with plenty of bamboos. Wild mango is common. Mahua is conspicuously absent in the hill region but it is plentiful in the plain area. Wild life has declined to a very low level.

3.2.2 House hold and community

Abujhmria houses are of purely temporary type. Once constructed, they are never repaired, the idea being that in any case a new one is to be built after about five years, so why to repair. Nevertheless, Abujhmria houses have definite design with five basic parts viz. living room in which outsiders can come and go (algi), kitchen in which house wife also sleeps (alpanji), husbands sleeping room (agha), sanctum sanctorum (out sides not allowed to enter this room) and vende kurma or the menstruation shed (Fig. 3.2). The husbands are normally confined to algi and agha while the women’s domain is all over but she also confines herself to algi and alpanji. House hold possessions are meager. Husking rods, gourd pots, bark rain coat, rain hat, and rain shield, axe, bows and arrows, fish traps, blankets and some bottles are all that one can see in Abujhmria house in addition to utensils and sleeping mats.
3.2.3 Physical characteristics

Abujhmarias have well developed bodies. The men do not carry load on their head but the women do so. The men carry load over their shoulders. Both men and women are great walkers, walking 30 to 40 kilometers up and down the hills is no problem to them. Abujhmarias are scantly clothed. They do not wash after defecation. They rarely take bath. Abujhmarias are light hearted people always joking and laughing. They are a contented people who always live in the present and do not care to remember the past or to hope for future. They are extremely shy and timid as far as outsiders are concerned. Everybody pays equal regard to the protection of other’s possessions with strong feeling of cooperation.

3.2.4 Food and drinks

Abujhmarias are omnivorous. The grain from penda fields, supplemented by large number of roots, tubers, leaves, shoots, seeds, fruits, animals and jungle creatures locally available constitute the food ingredients. Many of the items eaten are yet to be identified and known.

3.2.5 Social organization

Abujhmaria clan is divided into two groups viz dadabhai and akomama. The term dadabhai made up of two words dada and bhai both of which mean
brother. Similarly, the term *akomama* is also made up of two words *ako* and *mama*. Ako meaning brother and mama meaning mother’s brother. These words are also respectively used for daughter and daughter’s son. Marriage cannot take place between clans which are dadabhai to each other. Every Abujhmar village has a *patel*-spokesman for the whole village when dealing with the outsiders and authorities. But the religious head man is the *Kasyeg Gaita* who gets a special seat of honour during meeting held to decide shifting of the cultivation sites etc. The social organization of the Abujhmarias, the local group has very important place by the traditional standards of the tribe. The local group encourages its members by according social approval. It keeps an eye that laws, customs and traditions are not breached by the members. Abujhmaria villages also have dormitories (*Ghotul*) but only open to boys (Fig.3.3). This is so because the Abujhmaria villages are largely uni-clan ones and boys and girls of the same clan are considered as brothers and sisters. The Abujhmarias feel proud for their *ghotuls* as the unit serve as rest house for outsiders, to keep the younger members of the tribe organized and available for common assembly purposes. Various traditional musical instruments like drum, flute are kept in *ghotul* as these are used during recreation. A fire is always there in front of the *ghotuls* and different families take burning sticks from here to light their hearth.

![Ghotul](image)

Fig. 3.3  Ghotul a multipurpose structure used for Abujhmaria boys, rest house for outsiders and as a place for community functions and meetings.
3.2.6 The individual

Individual have no place in Abujhmaria society. There is collective and communal life – the land belongs to the village, decisions are taken collectively, religious ceremonies are celebrated collectively and economic pursuits like penda cultivation, threshing of harvest, hunting and fishing are all collective operation. If at all any unit could be thought of below the village, it would be the family and not any individual. Even no one can give his daughter anything in marriage unless the village approves of it. The Abujhmaria women are regarded in high esteem but still her position is markedly inferior to that of man. Tribal custom has imposed upon her a number of economic and socio-religious disabilities viz. she cannot sleep on cot, cannot participate in religious ceremonies, cannot inherit property etc.

3.2.7 Other social characteristics

Most of the deliveries take place in the menstruation hut. In delivering a child, the woman is not assisted by any body. She has to even cut the umbilical cord. If the child is still-born, she has herself to dispose it off. She returns to the house only after the naval cord of the child dries off. In Abujhmaria society, monogamy is the general rule but an appreciable proportion of men have more than one wife. This situation is just a means to accommodate the excess women in the society itself. Junior Levirate is the general rule that a widow becomes the wife of the deceased’s younger brother. Divorce is theoretically possible but in practice there are no such cases.

3.2.8 General health

Casual observations tell upon the Abujhmarias’ health as good. But closer look speaks upon otherwise. The youth of Abujhmaria is very short lived. The youth ages very fast. Conjunctivitis is widespread in Abujhmarrh. Also there is appreciable problem of men suffering from hydrocele. Abujmarh continues to remain one of the few pockets in the country where PF malaria still survives. This *plasmodium falciparum* (PF) is a certain killer.
3.3 **Sampling Procedure**

The location of the study area was selected purposely where as the respondents were selected randomly for collection of data. Random purposive sampling procedure was used to select the respondents in the present study.

3.3.1 **Selection of the sample**

In order to obtain a true representative sample, the sample should be selected at random (Swaminathan, 1974). For the present study 300 respondents from four villages of the Orchha Gram sabha were selected. According to 2001 census, the population of Orchha Gram sabha was 2539 residing in four villages (Appendix–V). The respondents in their families covered in the survey constituted infants, children, adolescents, adults and elderly people. As suggested by Jelliffe, the adult subjects were divided into 3 age groups viz. Young adult (18-35 years), Adults (36 to 55 years) and Old adults (>55 years). The subjects surveyed were homogenous in their character and way of living. On the whole, selection of sample was done on the basis of random purposive sampling for which heads of the families were interviewed. Collection of forest produce and agricultural produce is daily routine of Abujhmaria, therefore the head of the families were available for interview only in the morning. Few of them were interviewed in weekly market.

3.3.2 **Interaction with selected respondents**

The area selected was a prohibited place for visitors. Special permission from District collector was taken to visit the area. Local officers of Child development project, ICDS and ‘angawadi’ were approached for assistance. The purpose of the study was explained to the local officers and the interaction with the respondents was done in association with them (Fig. 3.4). The Abujhmarias usually do not share their knowledge with outsiders but the task could be accomplished with the help of local officers who were acquainted with them.
3.4 Socio economic scenario

All the families practice Shifting cultivation (penda). About 6000 hectare land is under settled agriculture. There engagement with food related activities is shown in Fig. 3.5. Domestication of cattle wealth with Abujhmarias has taken place. However, they do not milk the cows and also do not consume milk. They are great experts in bamboo work. Big basketry, large baskets, dancing shields, sleeping mats, rain hats, basketry plates and rain shields are prepared and sold in markets. Phool bahari grass grows in abundantly and broom making is quite popular for sale in the area. Rope making from firass (mawra) for own use and sale is common. Principal source of income of the Abujhmarias are - sale or barter of forest produce like chironji, tendu, gum, dried mango slices, tamarind, cocoon, phool bahari, baskets and rope, sale or barter of hill pulses (Arhar) and sarson, kulthi, bajra and kosra, sale of cattle etc. The people are not yet conversant with money economy.
Cutting of bamboo and other vegetation
Burning of cut vegetation for Penda
Sowing of different crops
Sowing of crops and weeding
Harvesting of crops begins
Weeding and harvesting
Harvesting, threshing and storage
Collection of Forest Produce
Harvesting of millets and pulses
Sowed trees start for clearing land
Cutting of trees starts for clearing land
Collection of Forest Produce
Fig. 3.5 Annual Economic Activity Chart of Abujhmaria Tribe
Pretested schedule prescribed by NIN, Hyderabad was used for conducting socio-economic survey. Some modification in the schedule was made to suit the local need (Appendix-II). The information was collected through oral questionnaire.

3. 5 Tools and Techniques of data Collection

Standard methods were followed for collection of data from the study area. Data was collected during the year 2003-04. The techniques and procedures on different aspects are presented as follows:

3.5.1 Diet survey

For conducting diet survey, the schedule described by ICMR with some modification was used. (Appendix-III) The food intake for three consecutive days was assessed by oral questionnaire (24 hour recall), using standard measures. Raw food ingredients were measured by NIN standard cups for determining the food consumption. The weights were taken for three consecutive days in each house hold. The average consumption per consumption unit was finally analysed from food intake and nutritive value was calculated using food tables. Comparison with recommended allowances of ICMR (1990) was then made.

The intake in terms of consumption unit or per person per day is expressed as follows:

\[
\text{Intake per consumption unit /day} = \frac{\text{Total raw amount of each food stuff}}{\text{Total consumption units of the family} \times \text{Total number of days of survey}}
\]

\[
\text{Intake per person per day} = \frac{\text{Total raw quantity of each food stuff}}{\text{Total numbers of members in the family who par take the meal} \times \text{Total number of days of survey}}
\]
3.5.2 Health care practices

Tribals believe that most of the diseases are caused by evil spirits, anger of clan Gods, Bongas and breach of taboos together with other ideas namely, humoral, hot and cold theories. As per their concepts most of the diseases of children and women are caused by evil spirits. Epidemic diseases are caused by the anger of clan Gods and Bongas, and personal diseases are caused by breach of taboos whereas venerable diseases are caused by breach of sex taboos. Abujhmarias also follow traditional practice of health care through local medicine man who is believed to have super-natural power. They also collect and use variety of plant leaves, fruits, seeds, nuts, roots, bark etc, that have great medicinal value and are used in curing common ailments.

3.5.3 Collection of rare food

Necessities of the Abujhmarias tribe are very few, mostly limited to the essential need of life. Abujhmarias are omnivorous tribe, eating almost everything which could be eaten. There are a number of food items consumed by the Abujhmarias which are rare. Some of the rare / less familiar foods consumed by Abujhmarias were collected.

3.6 Food analysis

The Abujhmarias due to their low economic status and non availability of edible food stuffs, sometimes consume food items that are not generally used by other human races. Such food items are not known to the people in general. Such rare food items were analysed for their nutritive value.

3.6.1 Estimation of Energy

Energy was calculated by sum of physiological energy values of carbohydrate, fat and protein.

3.6.2 Estimation of protein

Protein content was estimated by Micro-Kjeldahl method as described by AOAC (1980)
Principle

The estimation of nitrogen was done by Micro-Kjeldahl method. The nitrogen in protein or any organic material was converted to ammonium sulphate by sulphuric acid during digestion. This on steam distillation liberates ammonia which was collected in boric acid solution and titrated against standard acid. Since 1ml of 0.1 N acid was equivalent to 1.401 mg N, calculation was made to arrive at the nitrogen content of the sample.

Reagent

1. Catalyst mixture: K$_2$SO$_4$ (100g), CuSO$_4$(20 g)
2. Sodium Hydroxide (40%)
3. Sodium Hydroxide (0.1N)
4. Sulphuric acid (0.1N)
5. Mixed indicator: Bromocresol green (0.099g), Methyl red indicator (0.066g) dissolved in 100ml of ethanol.
6. Boric acid (2%)

Procedure

Sample, (0.5g) was taken in digestion tube. 10ml of concentrated H$_2$SO$_4$ and 2g of digestion mixture (100g of K$_2$SO$_4$ and 20g of CuSO$_4$, 5 H$_2$O) were added and the material was digested in the digestion chamber. The digested material was transferred to 10ml volumetric flask and volume made up using distilled water. A 10ml aliquot was pipetted into distillation apparatus and then steam distilled in presence of 10ml of 40% NaOH solution (Kjeldahl method).

The distilled ammonia was absorbed in 10ml of boric acid (2%) containing mixing indicator. The nitrogen was determined by titrating the collected distillate against H$_2$SO$_4$. Total protein was estimated by total nitrogen value multiplying with 6.25 which gives crude protein content that includes non protein nitrogen.

3.6.3 Estimation of fat

Fat content was estimated in dry material by using soxhlet Method (Sadasivam and Manikam).
Principle
Fat is extracted with petroleum ether from the dried sample. The solvent is removed by evaporation and the residue of fat is weighed.

Reagents
Petroleum ether, boiling range 40-60°C.

Procedure
5 g well ground dried sample was taken in an extraction thimble. The thimble was placed in the extractor and was connected to a weighed flask containing 100ml petroleum ether. The extractor was connected to a reflux condenser. The sample was extracted under reflux for 5-6 hours. The petroleum ether extract was evaporated to dryness. The flask containing the fat residue was dried in an air oven at 100°C for 5 minutes, then cooled in a desiccator and then weighed.

3.6.4 Estimation of carbohydrates
The total percent of the carbohydrates was determined by spectrophotometer method by using anthron.

Principle
Carbohydrates are first hydrolysed into simple sugars using dilute hydrochloric acid. In hot acidic medium glucose is dehydrated to hydroxymethyl furfural. This compound with anthron forms a green coloured product with an absorption maximum at 630 nm.

Reagent
1. Hydrochloric acid (2.5N)
2. Anthron reagent: 200mg anthrone dissolved in 100ml of ice cold 95% H₂SO₄. This reagent was always prepared fresh before use.
3. Standard glucose: (a) Stock- 100mg glucose was dissolved in 100ml distilled water (b) Working standard – 10ml of stock was diluted to 100ml with distilled water. It was stored in refrigerator.
 Procedure

100mg of the sample was weighed into a boiling tube. It was hydrolysed by keeping it in boiling water for three hours with 5ml of 2.5 N HCl and cooled to room temperature.

It was neutralized with solid carbonate until the effervescence ceased. The volume was made up to 100 ml and centrifuged. The supernatant was collected and 0.1 ml aliquots was taken for analysis and volume was made 2 ml as standard. Glucose solution (Conc. 1x 10^{-2}g / ml) standards were prepared by taking 0, 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4, 1.6, 1.8 and 2ml of the working standard. ‘O’ serves as blank. The volume was made up to 2 ml in all the tubes including the sample tubes by adding distilled water. Then 4ml of anthrone reagent was added in samples and standard solution then heated for eight minutes in a boiling water bath. It was cooled rapidly, and the green colour was read at 630nm.

A standard graph was drawn by plotting concentration of the standard on the X- axis versus absorbance on the Y- axis. From the graph the percent of carbohydrate present in the sample was calculated.

3.6.5 Estimation of moisture

Moisture content was estimated by drying the material in an oven at 100 to 105\(^\circ\) C and weighing it before and after drying (NIN, 1983)

 Procedure

About 10 g of material was weighed into a weighed moisture box and dried in an oven at 100-105\(^\circ\)C and cooled in a desiccator. The process of heating and cooling was repeated till a constant weight was achieved. The moisture was calculated as follows:

\[
\text{Moisture} \% = \frac{\text{Initial weight} - \text{final weight}}{\text{Weight of the sample}} \times 100
\]

3.6.6 Estimation of crude fiber (neutral detergent fiber)

The estimation of crude fiber was done by the method of Goering and van Soest (1970).
Reagents

1. Neutral detergent solution was prepared by dissolving 18.61 g di-sodium ethylene diamine tetracetate dehydrate (EDTA) and 6.81 g sodium borate dehydrate in about 500 ml of water by heating on a boiling water bath. Then 30 g sodium lauryl sulphate dissolved in about 200 ml of hot distilled water was added. To this 4.56 g anhydrous di-sodium hydrogen phosphate, dissolved in about 100 ml hot distilled water was mixed. After cooling, 10 ml of 2-ethoxyethanol was added to the mixture and volume was made to one litre.

2. Acetone

3. Sodium sulphite

Procedure:

500 mg dried material was refluxed with 100 ml of neutral detergent solution containing 0.5 g sodium sulphite in a 600 ml beaker without spout for one hour on a refluxing apparatus. The extracted material was filtered through a glass crucible (GI) under vacuum. Any residue left in the beaker was transferred to the crucible with hot distilled water and filtered again. This washing procedure was repeated twice and final washing was done with acetone till the filtrate was free from colour. The crucibles were kept in oven maintaining 100°C for overnight, cooled in a dessicator and weighed again. The gain in weight of the crucible was expressed as neutral detergent fibre.

3.6.7 Estimation of Total Ash

Principle

Ash is the inorganic or mineral component of the sample left after complete ignition of the sample at 600°C in muffle furnace.

Procedure

About 5-10 g of the sample was weighed accurately into a tarred porcelain crucible (which was previously heated to about 600°C and cooled). The crucible was placed on a clay pipe triangle and heated, first over a low flame till all the material was completely charred, followed by the heating in a muffle furnace for about 3-5 hours at about 600°C. It was then cooled in a dessicator and weighed. To ensure completion of ashing, the crucible was
again heated in the muffle furnace for 1/2 hour, cooled and weighed. This procedure was repeated till the two consecutive weights arrived were the same and the ash was almost white or greyish white in colour.

3.6.8 Estimation of Ascorbic acid

Vitamin C in the dietary samples was estimated using 2, 6-dichloroophenol endophenol dye (NIN 1983).

Principle:

The blue colour produced by the reduction of 2, 6-dichloroophenol indophenol by ascorbic acid is estimated colorimetrically.

Reagent:

1. Acetate buffer, pH 4.0 : 300 g of anhydrous sodium acetate dissolved in 700 ml of water and 1 litre of glacial acetic acid were mixed.
2. Dye solution: 25 mg of sodium salt of 2, 6-dichloroophenol indophenol is dissolved in distilled water and it was made up to 200 ml.
3. 6% Metaphosphoric acid (H\textsubscript{3}PO\textsubscript{3})
4. Ascorbic acid standard (1 mg / ml)

Procedure

5 g sample was blended with 6 % metaphosphoric acid to make 50 ml and 5 ml of the slurry was further diluted to 50 ml.

2.5 ml filtrate was placed in a 50 ml separating funnel (A). The same amount of metaphosphoric acid was taken in two more separating funnels (B) and (C). Funnel ‘B’ served as the dye blank and to funnel ‘C’ that served as a standard, was added with 0.1 ml (equivalent to 0.1 ascorbic acid) of the ascorbic acid standard solution. An amount of acetate buffer equal to the volume of the extract taken was then added to all three funnels, followed by 2 ml of the dye solution. Xylene 10 ml was also added quickly and the contents shaken for 6-10 seconds. After the layers separate, the lower water layer was removed and the colour in the xylene extract was measured in a photoelectric colorimeter at 500 nm.
The ascorbic acid taken for reaction with the dye was equal to 0.1(b-a)/b-c mg. The ascorbic acid content of the material was then calculated by applying the necessary solution factors.

### 3.6.9 Estimation of minerals

The mineral content of the food stuffs was determined by atomic absorption spectrophotometer 4129.

**Principle**

Representative sample in a suitable liquid form is sprayed into the flame of an atomic absorption spectrophotometer and the absorption or emission of the mineral to be analysed is measured at the specific wavelength.

**Procedure**

1g of oven dried samples were digested by 10ml of diacid mixture HNO$_3$ : HClO$_4$ (Chapman and Pratt, 1961) using Gerhardt- Kjeldotherm unit. Volume of digested samples were made to 100ml for the estimation of Phosphorous, Potassium, Zinc, Iron, Copper and Manganese content.

P - Phosphorus content of samples were estimated by development of molybdovanadate yellow colour method (Jackson, 1958)

K - Potassium content in the digested samples were estimated by flame photometer.

Zn, Cu, Fe and Mn - Zinc, copper, iron and manganese content in the digested samples were determined by atomic absorption spectrophotometer 4129. (Lindsay and Norvell, 1978).

### 3.6.10 Estimation of Calcium

**Principle**

Calcium is precipitated in acidic medium as insoluble calcium oxalate by adding saturated ammonium oxalate solution. The precipitate is dissolved in dilute sulphuric acid (1:9), heated and the oxalic acid thus released is titrated against standard potassium permanganate solution in warm condition ($60^\circ$ C) to get the calcium content of the sample.

**Reagents**

1. Saturated solution of ammonium oxalate.
2. Ammonium hydroxide solution (1: 4)
3. Hydrochloric acid solution ( 5%)
4. Sulphuric acid solution (1:9)
5. N/10 KMnO₄.
6. Methyl red indicator (Dissolve 0.1 g of methyl red in 100ml of 95% alcohol).
7. Calcium chloride solution.

Procedure

In 100 ml beaker, 10 ml of hydrochloric acid extract was taken. To it 2-3 drops of methyle red indicator was added. The extract mixed with methyle red indicator was heated to boiling point and cooled. To this 10 ml of saturated ammonium oxalate solution was added slowly with constant stirring until the precipitate become coarsely granular. The coarsely granular precipitate was again heated to boiling point, cooled and to it ammonium hydroxide (1:4) was added until the colour became faint pink. It was allowed to stand overnight to settle the precipitate. It was filtered through Whatman filter paper. No. 40 and washed with hot water, till the precipitate was free from soluble oxalates. Break the filter point of the filter paper with glass rod and washed the precipitate into the beaker in which calcium precipitated. It was dissolved in about 10 ml of dilute sulphuric acid (1:9). Heated to about 60°C and titrated against N/KMNO₄. Faint pink colour persisting for at least 30 seconds indicated that the titration is complete.

3.7 Statistical Techniques

The data was analysed by frequency, percentage, mean, standard deviation, chi square based on the nature of data and the type of information required. Statistical analysis was done using SPSS Computer software version 13.0 (Demo version)

Percentage – Simple comparisons were made on the basis of percentage calculation.

Chi square test: This test was used for determining the association between related variables. Knowledge regarding Health and Hygiene, immunization etc. were analysed by using this test.