Different methodologies have been applied for the study of the organic remains obtained from the rock samples. Various techniques and uses of different chemicals have been proposed by various authors in extraction of fossil palynomorphs from rock samples:

- Material collection
- Extraction of palynofossils
- Preparation of slides
- Microscopic Examination
- Diagrams
- Photography

**Materials:**

The materials of the present thesis have been collected from South-east of Inspection Bungalow, 1/4 Km, Mawsynram road Section (In between Phunsoryng village & Lower Fuxura village) ½ Km north-east from the village road section, Tlonplam village (along Mawsynram –Balat Road Section 15Km. south of Mawsynram Inspection Bunglow, Shillong –Cherrapungi road Section. Near 5 Km. milestone in Cherrapungi-Mahadeo Road Meghalaya, northeastern India.

Three field collection programme were conducted during September 1998, March 2000 and March, 2001 .Details of the collected samples including the litho logy and stratigraphic position are given in Table No 2

**Extraction of palynomorphs**

The samples taken for present investigation show a wide variation in their lithological characters ranging from clay to conglomerate including sandstone, shale, coal and limestone (refer Table No.2). As such different methodologies have been employed to extract the organic matters from the samples. However, the extraction technique proposed by Gray (1965) has been followed with required alteration and modification during the course of the work.

**Cleaning and Crushing**

Cleaning of the samples is very much essential to avoid surface contamination. The samples have been cleaned after scraping the outer surface and washing in the distilled water and then it has been allowed to dry in a pair of petridishes. Then it has been crushed into small pieces (about 1 cm in size) in mortar and pestle.
Use of Hydrochloric acid (HCl):
Common inorganic compound remain in the sedimentary rocks are carbonates like Calcite (CaCO₃), Dolomite [CaMg (CO₃)₂] Sulphates like Calcium sulphate and hydrides (CaSO₄) or gypsum (Cas₀₄, 2H₂O). These compounds are dissolved in HCl. To dissolve that unwanted compounds 10% aqueous solution of HCl has been used.

Use of Hydrofluoric acid (HF):
Silica is one of the most common materials in the collected samples. Silica may remain in the samples in the form of silicates like quartz etc. which is easily broken down in hydrofluoric acid (HF). 50% HF has been used to disintegrate the siliceous matrix of the rock samples for 24-48 hours on each samples depending upon the degree of disintegration. In case of coal samples HF generally has not been used. Thereafter distilled water is mixed to dilute the concentration of HF and then HF is washed off by repeated centrifugation process.

Use of nitric acid (HNO₃):
Besides silicates, carbonates and sulphates several sulphide compounds are also common constituent of the collected sedimentary rock materials, such as pyrites (FeS₂), marcasite (FeS₂) etc. These compounds are soluble in concentrate HNO₃. Not only that but HNO₃ also help to dissolve the carbonate and sulphate compound. HNO₃ being an efficient oxidant oxidizes the organic materials in the samples. In case of coal the HNO₃ treatment [along with potassium chlorate (KClO₄)] has been carried out for 45 minutes to 24 hours, but in other cases the duration of the treatment period has been maintained in between 30-45 minutes depending upon the variability of the constituent of the rocks.

Use of potassium hydroxide (KOH):
After separating several inorganic chemical it is required to remove the humic matter from the samples. 10% (v/v) KOH has been used for 5-10 minutes for effective reaction to dissolve the humic matter.

Separation of organic matter using Potassium iodide (KI) and Cadmium iodide (CdI₂) solution:
366 gm CdI₂ and 332 gm KI have been taken and dissolved in water. The specific gravity of that solution has been adjusted at 2.0-2.2 by adding required distilled water to that solution. Then the solution has been filtered. Gray (1965) has suggested keeping the specific gravity of the solution at 2.5 whereas Hallick (1957) got the best result at a specific gravity 2. In this work a
compilation of these two has been done and the specific gravity has been kept at 2.0-2.2. That enables to get maximum amount of sporepollens through centrifugation - floatation technique, though few finer rock particles has been mixed with the supernatant. This unwanted material has been separated manually by panning method.

**Preparation of slides:**
The slides were prepared in two different methods:

1) To prepare the permanent slides fractional amount of macerated materials was taken directly from water and mixed with polyvinyle alcohol on cover glass and spread evenly. After proper drying the cover glasses were mounted on clean slides by using Canada balsam.

2) The slides were also prepared by using glycerine jelly after treating the samples with 10/glycerine. A fractional amount sample was then mixed with a little amount of glycerine jelly on a slide, spread evenly. Covered with a cover glass and then sealed with paraffin. The slides and the unused macerated materials are kept in the Paleobotany and Palynology laboratory, Department of Botany, University of Kalyani.

**Micoscopic Examination:**
The palynomorphs were examined in transmitted light with (Carl Zeiss Jenaval binocular microscope with 40x and 100x objectives and 15x eye piece using photomicrograph model no.) Leitz Laborlux S. binocular microscope with phaco lens.

**Diagrams:**
To find out comparative analysis of the frequency of microfloral composition of all specimens at random selection of slides with constant counting areas, the histograms were prepared.

**Photography:**
Photomicrography has been made by the wide field microscope with digital photo micrographic camera.
Flow Chart of Methodology Paleopalynological work

**FIELD**

**EXPOSURE**

- **OXIDATION**
  - By Centrifugation with Distilled water
  - For Coal

- **PIECES OF ROCKS TAKEN (ABOUT 500GM)**

- **TAKEN IN SAMPLING BAG, TAGGED AND NUMBERED WITH BRIEF DESCRIPTION**

- **PLOTTING THE TOPOGRAPH**

- **RECORDING THE FIELD BOOK**

- **SCHULZ SOLUTION (HNO₃+KClO₃) FOR COAL ONLY FOR OXIDATION**

- **APPLICATION OF HEAVY LIQUID (2.2 SP. GR.) FOR FLOTATION TECHNIQUE TO SEPARATE ORGANIC MATTER**

- **SUPERNATANT COLLECTED**

** METHODOLOGY**

- **EXTRACTION:**
  - **CLEANING**
  - **DRYING**
  - **CRUSHING IN MORTAR PESTLE UPTO 1 CM PIECES**
  - **POURING HCL (10% V/V) IF NECESSARY FOR 10 MINUTE TO REMOVE CARBONATES**
  - **POURING HF TO REMOVE SILICA FOR 20-48 HRS.**
  - **POURING HNO₃ (CONC) FOR OXIDATION FOR 30-50 MINUTE**
  - **POURING KOH (5% V/V) FOR 5-10 MINUTES TO REMOVE HUMIC MATTER**
  - **APPLICATION OF HEAVY LIQUID (2.2 SP. GR.) FOR FLOTATION TECHNIQUE TO SEPARATE ORGANIC MATTER**

- **CLEANING**

- **WASHING**

- **WASHING**

- **WASHING**

- **WASHING**

- **WASHING**

- **WASHING**

- **WASHING**

**POURING DISTILLED WATER TO REDUCE SPECIFIC GRAVITY**

- **CENTRIFUGED**

- **WASHING**

- **PALYNOMORPHS COLLECTED**

- **FIXATION IN POLYVENEY L ALCOHOL**

- **SPREADING ON SLIDE WITH GLYCERINE JELLY**

- **MOUNTED IN CANADA BALSAM**

- **WAX SEALING**

- **OBSERVATION**

- **TAXONOMIC STUDY**

- **PHOTOMICROGRAPHY**

- **CALCULATION**

- **CONCLUSION**