MATERIAL AND METHOD
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The present study was carried out in the Department of Obstetrics and Gynaecology, M.L.B. Medical College, Jhansi over a period of one year.

Patients were selected from the out patient department and ward of Department of Obstetrics and Gynaecology of M.L.B. Medical College, Jhansi.

Categorization of cases: Cases were studied into two groups:

1- Control Group:

The control group comprised all the cases attending our out patient or admitted in ward with normal pregnancy of different trimesters and non pregnant healthy females of reproductive age group. Total number of cases included in this group was twenty five.

2- Study Group:

This group comprised women with previous history of pregnancy less in form of

- Abortions
- Congenital malformations
- Still births
- Preterm delivery

Total number of cases included in this group was ninety.
HISTORY:

A detailed history of each patient was taken followed by thorough general, systemic, and local examination as follows:

1- Name, age, OPD No. were recorded
2- Occupational status was considered in order to know socioeconomic status of the patient.
3- Education status of each patient was asked
4- Dietary history was asked whether vegetarian or non-vegetarian
5- History of any addiction e.g. smoking alcohol, tabacco was asked
6- History of present illness was elicited.
   An enquiry was made about the duration of pregnancy and onset of sign and symptoms in relation to period of amenorrhae if she come in antenatal period.
7- Past history of fever, lump in body, rashes, any eye complaints, cough, jaundice, diabetes, hypertension was enquired.
8- Family history of diabetes and hypertension was inquired
9- Obstetrical history, previous obstetrical history was taken.
   - Total number of time patient was conceived
   - Total number of full term pregnancy
- Abortions - duration of pregnancy at time of abortion, type of abortion.

- Congenital malformation - discovered by ultrasonography or by receiving product of conception or foetus.

- Premature delivery

- Still birth

- Number of living children and last child birth or abortion.

- Mode of deliveries, sex, weight of babies and condition of babies at birth and at present were noted.

10- Menstrual history - Date of last menstrual period was asked and expected date of delivery was calculated.

11- Drug history - Any treatment taken in past for any medical disease or for pregnancy losses.

EXAMINATION OF THE PATIENTS

1- **General Examination** - Thorough general examination was done with special attention to pallor, blood pressure, lymphadenopathy, temperature.

2- **Systemic Examination** - Brief systemic examination of cardiovascular system, respiratory system, central nervous system and of gastrointestinal system was done. This was to exclude any systemic disease.
3- **Obstetrical Examination**: Thorough obstetrical examination was done as fundal height, lie presentation and fetal heart rate.

4- **Per vaginal Examination**: It was done whenever necessary as for confirmation of pregnancy in first trimester and when patient complained of pain during pregnancy.

**INVESTIGATIONS**: 

During the first visit the following investigations was done:

1- **Haemoglobin percentage estimation** was done using Sahili's method.

2- **Total leucocyte count, differential leucocyte count and erythrocyte sedimentation rate** was done to diagnose any infection.

3- **ABO and Rh grouping** was done because ABO, Rh incompatibility is one of important causes of BOH.

4- **VDRL** was done in each patient.

5- **Complete urine examination routine for albumin and sugar and microscopic for pus cell, RBC or any cast.**

6- **Fasting and post prandial blood sugar examination** was done in each patient.

7- **Ultrasound examination of lower abdomen** was done in each patient.
- To see for any congenital malformation of uterus
e.g. double uterus, septate uterus etc.
- To know about any uterine disease e.g. fibroid uterus
- To know gestational age of foetus

We can exclude gross congenital malformation of foetus by ultrasonographic examination, for foetal well being.

Collection of the sample: 3 ml of venous blood was taken in a dry vial with autoclaved syringe and needle, blood was allowed to clot then tube was related between palm for 2-3 times and clot separated. Then test tube was placed in a incubator in standing position for half an hour. Test tube is then centrifused and serum was seperated, test of Ig G antibody for toxoplasma gondii was done from the seperated serum.

Sample preparation:

Mix patient's serum - 10 ul
Sample dilution buffer - 1 ml

Serum sample may be diluted at the time of use and stored at 2-8°C before testing for a day.

Methods for detection if Ig G antibody for toxoplasmosis

- Indirect fluorescent antibody test (IFAT)
- Passive hemagglutination test (PHA)
- Methylene blue dye test (MBD)
- Complement fixation test (CF)
- Enzyme linked immuno sorbet assay test (ELISA)

Method used in this study was ELISA

Principle: Toxo Ig G is a sandwich enzyme linked immunosorbent assay for qualitative determination of Ig G antibodies to toxoplasma gondii in serum.

Immunosorbent assay means that antigen in antibody detection tests are attached to a solid phase (Sorbent). Enzymes are linked to antibodies and react with a substrate indicating the immunological reaction through production of colour.

Procedure: Solid phase ELISA test consists of microtitre strips coated with toxoplasma antigen. If the patient's serum has the relevant specific toxoplasma gondii antibodies they bind to toxoplasma antigen an solid phase. After washing the bound antibodies are sandwiched using HRPO labeled anti-human Ig G conjugate resulting in formation of toxoantigen human Ig G HRPO labeled antihuman Ig G conjugate complex. The unbound conjugate is removed by washing and the enzyme linked sandwich complex is revealed by chromogenic substrate the intensity of the colour developed is directly proportional to amount of Ig G toxoplasma gondii antibodies in the serum.
After stopping the reaction with stopping solution absorbance is measured at 450 nm using an ELISA reader. Results of patients samples are obtained by calculation/comparison using the negative and positive controls.

Calculations:

1- Calculate the mean absorbance reading of negative control (MNC).

2- Calculate the mean absorbance reading of cut off control (MCC).

3- Calculate the mean absorbance reading of medium positive control (MMPC).

The test sample with absorbance values greater than or equal to the MCC are considered positive for Ig G anti toxoplasma gondii antibodies. The test samples with absorbance values less than MCC are considered negative.

If an ELISA reader is not available a visual interpretation of result is possible. A specimen can be considered positive if the colour intensity in the sample well is equal to or stronger than the colour intensity in the cut off control wells.