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Original Article

Paediatrics Section

# Role of Autopsy and Genetic Testing in the Diagnosis of Perinatal Deaths due to Congenital Anomalies: A Cross-sectional Study

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## ABSTRACT

**Introduction:** Foetal autopsy is one of the primary modalities for establishing the underlying causative aetiology in congenitally anomalous foetuses. Genetic aetiology is the cause in at least half of the foetuses. A correlative approach including dysmorphological evaluation, histopathology, imaging studies, and genetic testing plays a significant role in establishing the final diagnosis and management of such cases.

**Aim:** To investigate the cause of perinatal death through a correlative approach involving Ultrasonography (USG), cytogenetic analysis, and autopsy in congenitally malformed neonates.

**Materials and Methods:** This cross-sectional study was conducted in the Department of Pathology and Obstetrics and Gynaecology at SCB Medical College, Cuttack, Odisha, India, from June 2018 to June 2020. The study included 19 congenitally malformed foetuses/aborted foetuses/neonates not compatible with life, born in the labour room. Written informed consent was obtained, and cord/cardiac blood/soft tissue samples from congenitally anomalous foetuses were collected in a heparinised vial/sterile container during delivery and sent for cytogenetic testing. The deceased foetuses were weighed, external abnormalities and anthropometry were recorded, and then they were preserved in 10% formalin. Autopsies were performed using Virchow's method. All internal abnormalities were recorded, and sections were sent for Histopathological (HP) study. All the data was analysed using the Statistical Package for Social Sciences (SPSS) software version 20.0.

**Results:** The study included a total of 19 cases, including 11 (57.8%) cases of Intrauterine Deaths (IUD). The majority were males 8 (42%), in the gestational age group of 20-25 weeks (36.7%). Maternal age ranged from 21 to 30 years, with the majority being nine cases (47.3%), and 13 cases (68.3%) without antenatal check-ups. The study comprised 6 cases (31.57%) of genetic syndromes, including Trisomy 21, 13, 18, and Monosomy X, 5 cases (26.3%) of musculoskeletal defects such as Meckel Gruber syndrome, Heterotaxy syndrome, and Thanatophoric dysplasia, and 5 cases (26.3%) of neural tube defects.

**Conclusion:** Autopsy determined the cause of death in 90% of the cases and, when combined with genetic analysis, established the syndromic diagnosis. Autopsy findings can complement or modify the ultrasonographic findings. Therefore, perinatal autopsy should always be included in the management of deaths due to congenital anomalies.


**Keywords:** Cytogenetics, Down syndrome, Dysmorphology, Meckel gruber syndrome, Neural tube defects

## INTRODUCTION

Congenital anomalies can be defined as structural or functional anomalies (such as metabolic disorders) that occur during intrauterine life and can be identified prenatally, at birth, or sometimes only detected later in infancy, such as hearing defects [1]. This condition exists at birth and often before birth or develops during the first month of life (neonatal disease). Most malformations are the result of developmental defects. Congenital anomalies affect approximately 1 in 33 infants and result in approximately 3.2 million birth defect-related disabilities every year [1]. According to the World Health Organisation (WHO) in 2010, an estimated 270,000 deaths globally were attributable to congenital anomalies during the first 28 days of life, with neural tube defects being one of the most serious and common anomalies [2].

Foetal autopsy is one of the primary modalities for establishing the underlying causative aetiology in cases of morphologically abnormal pregnancies or intrauterine foetal demise. Various autopsy series in the past have demonstrated that postmortem evaluation provides additional findings or modifies the antenatal diagnosis, including Ultrasound (USG), in 20%-50% of cases [3-5]. Antenatal series have also shown that at least 50% of syndromic diagnoses are possible only after an autopsy [6]. Although USG can provide a fairly accurate diagnosis, autopsy examination of the deceased foetus for associated anomalies is essential to confirm the diagnosis.

Chromosomal abnormalities are reported in a significant proportion of pregnancy losses, suggesting an underlying genetic aetiology in many abnormal foetuses. Identifying the genetic cause in these cases brings an end to diverse diagnostic testing, eliminating the need for further costly tests [7].

Therefore, the present study was conducted to study the cause of perinatal death using a correlational approach, including USG, cytogenetic analysis, and autopsy in congenitally malformed deceased foetuses/neonates not compatible with life. The study also aimed to observe whether perinatal autopsy and cytogenetic testing are in concordance with the antenatal diagnosis.

## MATERIALS AND METHODS

This cross-sectional study was conducted at the Department of Pathology and Obstetrics and Gynaecology (O&G), SCB Medical College, Cuttack, Odisha, India, from January 2018 to January 2020. Ethical clearance was obtained from the Institutional Ethics Committee (reference no-923/14.10.19).

**Inclusion criteria:** All congenitally malformed dead foetuses/aborted foetuses/neonates not compatible with life, born in the labour room of the Obstetrics and Gynaecology Department during the study period, were included in the study.

**Exclusion criteria:** Cases with degenerated samples for genetic testing, autolysed foetuses, and those with unwilling parents/guardians were excluded from the study.

### Study Procedure

A proforma was used to record relevant data of the foetus and mother. Demographic factors of the mother, including age, socio-economic status, and consanguinity, were recorded. Detailed obstetric history, including the history of previous miscarriage/delivery of a congenitally malformed baby or Intrauterine Death (IUD), antenatal check-ups with a history of fever or infection, any drug intake, exposure to radiation, prenatal folic acid supplementation, and maternal co-morbidities, were recorded. Foetal parameters like gestational age, gender, birth status (live/stillborn/IUD), and details of congenital malformations, were included.

To detect the types of congenital malformations, antenatal ultrasound findings were noted in all six cases during antenatal check-ups and in 13 cases when the mothers attended the Obstetrics and Gynaecology (O&G) outpatient department and were admitted for obstetric procedures. Detailed external examinations were conducted to determine the location and type of anomalies. Anthropometric measurements such as Head Circumference (HC), Crown Heel Length (CHL), Crown Rump Length (CRL), Chest Circumference (CC), and Foot Length (FL) were noted and categorised as ≤1SD or ≥1SD based on a previous study by Archie JG et al., [8]. Certain anthropometric details were not included in cases of anencephaly, Down syndrome, and conjoined twins due to the impracticality or unreliable values they would provide.

Umbilical cords were examined for the presence of strictures, blood clots, and/or coiling. The placenta was dissected, and sections were taken for Histopathological (HP) study. Internal examination began with a modified "Y" shaped incision from behind each ear, down to the front of the chest, extending to the lower part of the sternum, then encircling the umbilicus on the left side and ending at the pubic symphysis. After opening the thoracic and abdominal cavities, the organs were individually removed (using Virchow's technique) and observed for any anomalies [9]. The weights of relevant (abnormal) organs were recorded, and sections were taken from them for HP examination.

Antenatal ultrasonographic diagnoses were compared with postnatal autopsy findings, and the association of congenital malformations with genetic parameters was analysed. Genetic analyses were conducted in registered labouratories. Cord blood samples were collected in heparinised vials during delivery, and if cord blood collection was not possible, cardiac puncture blood samples were taken and sent for karyotyping. Soft tissue samples from the foetuses, obtained from the ear lobule, plantar aspect of the leg, or anterior thigh, were placed in sterile containers and sent for Flourescence In-situ Hybridisation (FISH) study.

## STATISTICAL ANALYSIS

All the data was entered into Microsoft excel version 2203 and analysed using the Statistical Package for Social Sciences (SPSS) software version 20.0. The categorical variables were described as percentages. The numerical variables were compared using a student's t-test and a Chi-square test. A p-value of less than 0.05 was considered statistically significant.

## RESULTS

In the present study, congenitally anomalous foetuses, including stillborn, Intrauterine Demise (IUD), and neonates not compatible with life, were included. Autopsies were carried out, along with genetic testing and antenatal Ultrasound (USG). The Gestational Age (GA) in the study ranged from 19 to 35 weeks, with a minimum age of 19 weeks. The majority of cases (7, 36.8%) were between 20-25 weeks of gestation, and 8 (42%) were male, with 5 (26%) cases of ambiguous genitalia. Additionally, the majority of foetuses experienced intrauterine demise, followed by live-born neonates not compatible with life [Table/Fig-1].

| Demographic factors | | No. of cases | Percentage (%) |
|---|---|---|---|
| Gestational age (in weeks) | <19 | 03 | 15.7 |
| | 20-25 | 07 | 36.8 |
| | 26-30 | 06 | 31.5 |
| | 31-35 | 03 | 15.7 |
| Gender | Male | 08 | 42.1 |
| | Female | 06 | 31.5 |
| | Ambiguous genitalia | 05 | 26.3 |
| Type of birth | Still/IUD | 11 | 57.8 |
| | MTP | 02 | 10.5 |
| | Live born not compatible with life | 06 | 31.5 |

**[Table/Fig-1]:** Foetal demographic factor.
IUD: Intrauterine death; MTP: Medical termination of pregnancy

The age of the majority of mothers ranged between 21-30 years. Out of 19 cases, only 6 (31.5%) mothers went for antenatal check-ups, while the remaining 13 cases underwent USG when the mothers attended the O&G Outpatient Department at SCB Medical College and Hospital. Previous history of anomalous foetuses was present in 2 (10.5%) cases, and 5 (26.3%) mothers had a history of spontaneous abortion. In the studied population, only 4 (21%) mothers took folic acid supplementation. Many mothers had medical disorders complicating their pregnancy, with the maximum number (14, 73.6%) suffering from anaemia, followed by pregnancy-induced hypertension and hypothyroidism in 5 (26.3%) cases each [Table/Fig-2].

| Entities | | No. of cases | % |
|---|---|---|---|
| Age group (in years) | 18-20 | 3 | 15.7 |
| | 21-30 | 9 | 47.3 |
| | 31-40 | 7 | 36.8 |
| Previous pregnancy outcomes and consanguinity | Spontaneous abortion | 5 | 26.3 |
| | IUD | 2 | 10.5 |
| | Congenital malformation | 2 | 10.5 |
| | Consanguinity | 1 | 5.3 |
| Antenatal check-ups | Yes | 6 | 31.5 |
| | No | 13 | 68.4 |

| | | | |
|---|---|---|---|
| Socio-economic status | Lower | 6 | 31.5 |
| | Upper lower | 4 | 21 |
| | Lower middle | 4 | 21 |
| | Upper middle | 3 | 15.7 |
| | Upper | 2 | 10.5 |
| Folic acid, fever, radiation and drugs | Folic acid supplementation | 4 | 21 |
| | Fever | 6 | 31.5 |
| | Radiation | 0 | 0 |
| | Drugs | 4 | 21 |
| Co-morbid conditions | Anaemia | 14 | 73.6 |
| | Pregnancy induced hypertension | 5 | 26.3 |
| | Gestational Diabetes Mellitus (DM) | 1 | 5.3 |
| | Obesity | 2 | 10.5 |
| | Polyhydramnious | 5 | 26.3 |

**[Table/Fig-2]:** Distribution of maternal demographic factors.

Ultrasonographic findings in all the cases are provided in [Table/Fig-3]. On autopsy for organ-wise examination, the head and neck showed the maximum external dysmorphological findings in 9 (47.3%) cases, followed by the abdomen in 7 (26.3%) cases. Musculoskeletal deformities were found in 5 (26.3%) cases (perinatal X-ray showed smaller long bones), and the least abnormality was observed in the thorax and umbilical cord with placenta, each with 2 (10.5%) cases. The maximum number of internal deformities was found in the Cental Nervous System (CNS) with 7 (36.8%) cases, followed by the respiratory, Cardiovascular System (CVS), and Genitourinary System (GU), each with 5 (26.3%) cases [Table/Fig-4].

| Diagnosis | No. of cases | USG findings |
|---|---|---|
| Thanatophoric dysplasia | 3 | • Short and bowed foetal long bones, narrow chest, prominent forehead, wide spaced orbits, polyhydramnios<br>• Polyhydramnios, congenital anomaly<br>• Central craniosynostosis, strawberry sign, macrosomia, frontal bossing, flat face, lumbar hemivertibra, polyhydramnios |
| Down syndrome | 2 | • Left congenital diaphragmatic hernia, absent nasal bone, right axis deviation of heart, short long bones<br>• Cystic hygroma, CL, CP, upper and lower short limbs |
| Heterotaxy syndrome | 1 | Minimal foetal ascitis, scanty liquor amnii, small for date baby, thick placenta, small for date foetus |
| Turner syndrome | 1 | Cystic hygroma with foetal hydrops |
| Dextrocardia | 1 | Dextrocardia with situs inversus |
| Single umbilical artery | 1 | Absent ductus venosus, single umbilical artery, ventriculomegaly, CTEV |
| Edward syndrome | 2 | • Single umbilical artery, meningomyelocele, agenesis of right kidney, absent cisterna magna, TAPVC<br>• CLCP, single umbilical artery, absent intestinal loops |
| Conjoint twin with dingle heart | 1 | Parapagus twins with single heart |
| Anencephaly | 2 | • Anencephaly with polyhydramnios<br>• IUFD |
| Anencephaly with encephalocele | 1 | Omphalocele/gastrochiasis |
| Spina Bifida (SBf) | 2 | • Anencephaly/porencephaly<br>• Myelochiesis |
| Patau syndrome | 1 | -Alobar holoprosencephaly, foetal face with a common orbit (Cyclopia), facial dysmorphism |
| Mekel-gruber syndrome | 1 | Encephalocele with bilateral multicystic kidneys |
| Total | 19 | |

**[Table/Fig-3]:** USG findings in different cases.
CP: Cerebral palsy; CTEV: Congenital talipes equinovarus; TAPVC: Total anomalous pulmonary venous connections; IUFD: Intrauterine foetal demise

| Entities | | No. of cases | Percentage (%) |
|---|---|---|---|
| External dysmorphology | Head and neck | 09 | 47.3 |
| | Thorax | 02 | 10.5 |
| | Abdomen | 07 | 36.8 |
| | Musculoskeletal | 05 | 26.3 |
| | Placenta and umbilical cord | 02 | 10.5 |
| Internal dysmorphology | CNS | 7 | 36.8 |
| | CVS | 5 | 26.3 |
| | Respiratory | 5 | 26.3 |
| | GIT | 4 | 21 |
| | GU | 5 | 26.3 |
| | Haematolymphoid | 4 | 21 |

**[Table/Fig-4]:** System wise categorisation based on dysmorphological features on autopsy.

Congenital anomalies observed during the present study included six syndromes, comprising four types of cytogenetic syndromes: Trisomy 21 (Down syndrome), 18 (Edward syndrome), 13 (Patau syndrome), and Monosomy X (Turner syndrome) were diagnosed. The other two were Meckel Gruber syndrome and Heterotaxy syndrome. Additionally, 5 (26.3%) cases of neural tube defects and 3 (15.7%) cases of musculoskeletal defects were encountered [Table/Fig-5].

| Groups | Diagnosis | No. of cases (percentage) |
|---|---|---|
| Syndromes | Down syndrome | 2 (10.5) |
| | Heterotaxy syndrome | 1 (5.3) |
| | Turner syndrome | 1 (5.3) |
| | Edward syndrome | 2 (10.5) |
| | Patau syndrome | 1 (5.3) |
| | Meckel grover syndrome | 1 (5.3) |
| Neural tube defects | Anencephaly | 2 (10.5) |
| | Anencephaly with occipital Encephalocele | 1 (5.3) |
| | Myelochiesis | 1 (5.3) |
| | Meningomyelocele | 1 (5.2) |
| Musculoskeletal defects | Thanatophoric dysplasia | 3 (15.7) |
| Isolated anomalies | Dextrocardia | 1 (5.3) |
| | Single umbilical artery | 1 (5.3) |
| | Conjoint twin | 1 (5.3) |
| Total | | 19 |

**[Table/Fig-5]:** Distribution of different diagnostic groups based on autopsy and cytogenetic findings.

## Neural Tube Defects

This comprised the largest category of congenital anomalies and included three cases of anencephaly and two cases of Spina Bifida (SBf) [Table/Fig-5].

**Anencephaly (AN) [Table/Fig-6a,b]:** All three cases revealed similar external facies with absent calvaria, an extended neck, a

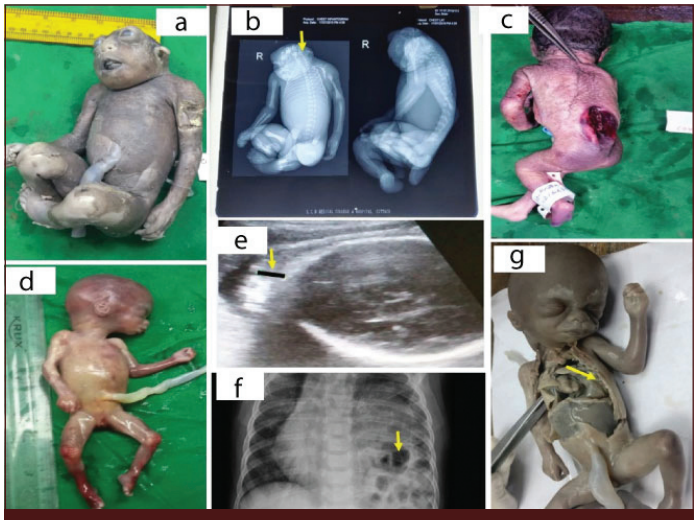
short forehead, and ocular protrusion. One case had an associated occipital encephalocele. The HC, CRL, and CHLs could not be recorded because of the absence of calvaria. CCs were on the lower side, and FLs were normal, as per the standard measurements. Internal examination revealed thymic hyperplasia in the first case, an enlarged midline liver and adrenal hypoplasia in the second case, and the third case showed distorted parenchymal tissue in the contents of the encephalocele and adrenal hypoplasia, which was confirmed by HP study.

**Spina Bifida (SB) [Table/Fig-6c]:** Both myelochiesis and meningomyelocele were present at the lumbar region. Apart from a slightly higher CRL and CHL, the rest of the anthropometric measurements were normal. Internal examination showed ventriculomegaly and a posterior vertebral column defect. Herniated neural elements were confirmed in the HP study.

## Syndromes

During the study period, eight documented cases were encountered, comprising six types of syndromes.

**Down Syndrome (DS) [Table/Fig-6d-g]:** Of the two cases of DS, one exhibited severe facial and skeletal dysmorphism, including cystic hygroma, unilateral cleft lip and palate, and a depressed nasal bridge. Both palms and feet originated directly from the chest wall and pelvis, respectively, with ambiguous external genitalia. Anthropometric examination recorded low HC, CC, CRL, and FL. The umbilical cord revealed two lumens, and HP study confirmed the presence of a single umbilical artery. Internal examination of the second case revealed left-sided diaphragmatic hernia with intestinal loops, right-sided heart deviation, and hypoplastic lungs.




**[Table/Fig-6]:** Anencephaly: a) Fetus with grossly absent calvaria, extended neck, ocular protrusion; b) X-ray picture-arrow showing absence of calvarium; c) Spina bifida (myelochiesis)- grossly open spina bifida; d-g) Down syndrome- d) gross picture showing depressed nasal bridge; e) USG showing increased nuchal translucency; f) X-ray showing multiple air spaces above diaphragm-arrow; g) autopsy showing depressed nasal bridge, micrognathia, diaphragmatic hernia of intestinal loops (arrow).

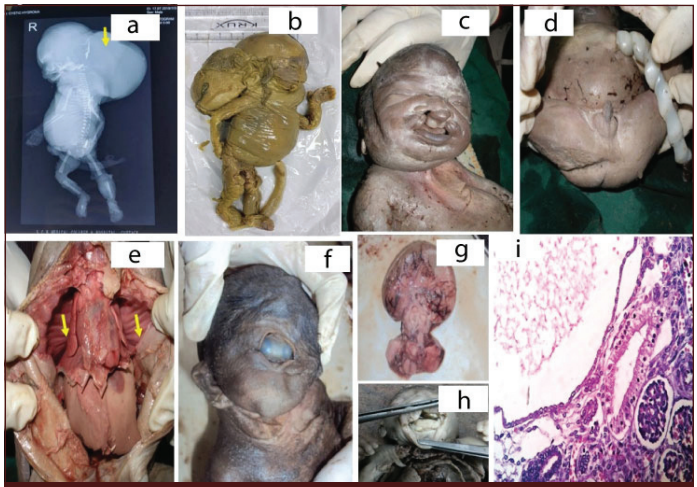
**Heterotaxy Syndrome (HS):** The present case did not present any external or anthropometric abnormalities. Internal examination revealed the presence of a trilobed left lung, a large liver, and the absence of thymus and spleen, which was confirmed by HP study.

**Turner Syndrome (TS) [Table/Fig-7a,b]:** The only case of Turner syndrome displayed cystic hygroma, a depressed nasal bridge, hypertelorism, a swollen abdomen, and ambiguous external genitalia. X-ray confirmed many of these findings. Anthropometry showed an increased CC, while the rest of the parameters were within the normal range. Internal anomalies included hypoplastic lungs and a fused kidney. The cut-section of the kidney exhibited multiple cysts, which was confirmed by HP study.

**Edward Syndrome (ES) [Table/Fig-7c-e]:** The present study included two cases of Edward syndrome. Externally, one case showed a lumbar meningomyelocele and a single umbilical artery, while the second case had bilateral cleft lip, cleft palate, and imperforate anus. CHL and CRLs were on the higher side. Internal examination of the first case revealed herniated material in the sac and ventriculomegaly, while the second case exhibited a collapsed lung, absence of spleen and intestine, right kidney agenesis, and streak gonads, which was confirmed by HP study. The karyotype showed trisomy 18.

**Patau Syndrome with Holoprosencephaly (PS with HP) [Table/Fig-7f,g]:** The foetus had a severe craniofacial defect with a single eyeball, a tubular nose-like structure, and an absence of the rest of the structures in the face. Anthropometry revealed HC, CRL, and CHL on the lower side of normal, while the rest of the parameters were normal. Autopsy of the foetus's skull revealed an alobar brain and a single optic nerve (severe type of holoprosencephaly-HP). Other viscera were normal externally and microscopically. Since it was associated with Patau syndrome, it was diagnosed as syndromic HP.

**Meckel Gruber Syndrome (MGS) [Table/Fig-7h,i]:** External examination showed occipital encephalocele, microcephaly, microphthalmia, orbital hypertelorism, low-set ears, cleft lip, and cleft palate, micrognathia, and a foot showing talipes equinovarus deformity. Apart from FL, which was normal, and CC, which was high, the rest of the parameters were on the lower side of normal. Internal examination showed distorted brain parenchyma in the encephalocele contents. The heart showed a bifid apex and atrial thrombi. The liver was enlarged with surface congestion. The cut surface of the kidney revealed multiple cysts. Microscopically, the liver showed fibrosis and lymphocytic infiltrations, and the kidney showed multiple cysts lined by flattened epithelial cells.




**[Table/Fig-7]:** a-b) Turner syndrome- a) X-ray picture -arrow indicates cystic hygroma; b) Gross picture of baby showing huge cystic hygroma, swollen abdomen; c-e) Edward syndrome showing c) Cleft lip, cleft palate; d) Imperforate anus, ambiguous genitalia; e) Autopsy revealing absence of right kidney, intestine and spleen (yellow arrow); f,g) Patau syndrome with Holoprosencephaly- f) Severe craniofacial defect with single eye; g) Alobar brain; h,i) Meckel Gruber syndrome- h) Clinical picture; i) HP section from kidney showing multiple cystic spaces lined by flattened epithelial cells, H&E x400.

## Musculoskeletal Defects

**Thanatophoric Dysplasia (TD) [Table/Fig-8a-c]:** All three cases of Thanatophoric dysplasia (dwarfism) exhibited similar external abnormalities, including craniofacial disproportion, frontal bossing,

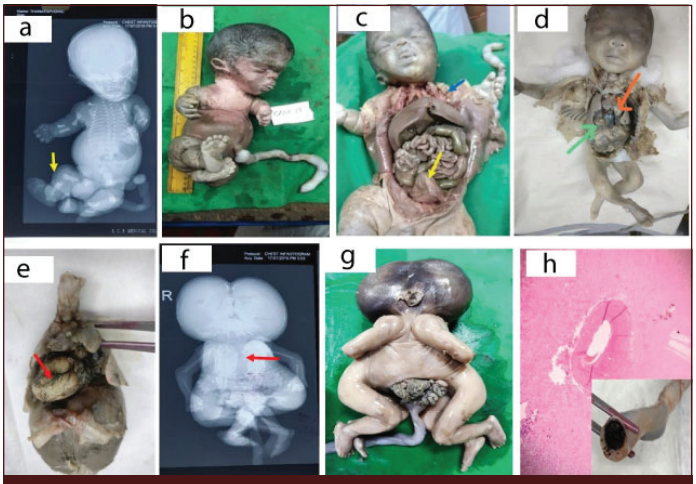
hypertelorism, depressed nasal bridge, narrow chest, protruding abdomen and genitalia, short symmetric upper limbs, and curved femur. Anthropometric evaluation indicated increased HC and CRL, as well as low CC, CHL, and FL. Internal examination revealed pulmonary hypoplasia and a large bladder. The musculoskeletal system showed short ribs and lumbar hemivertebra.

### Isolated Anomalies

**Dextrocardia (DXC) [Table/Fig-8d,e]:** This case exhibited unremarkable external features and anthropometry. Internal examination revealed right-sided heart placement and lung hypoplasia. After opening the thorax, the chest organs were removed en bloc to observe the relationship between the heart and lungs. Histologically, the organs were found to be normal.

**Conjoint twin [Table/Fig-8f,g]:** There was a single case of monochorionic-monoamniotic twin pregnancy. The foetuses shared two heads, one trunk, a single heart, four upper limbs, and four lower limbs [Table/Fig-3f,g]. The final diagnosis was dicephalic thoracoomphalopagus conjoined twins.

**Single Umbilical Artery (SUA) [Table/Fig-8h]:** External examination showed talipes equinovarus deformity and two vessels in the umbilical cord. Ventriculomegaly was found during the autopsy. CC was slightly raised, while the rest of the anthropometric parameters were within normal limits. Microscopically, the presence of a single umbilical artery was confirmed.




**[Table/Fig-8]:** Thanatophoric dysplasia- a)X-ray picture; arrow showing small long bone with curved femur; b) External abnormalities: Frontal bossing, depressed nasal bridge, short symmetric arm,narrow chest, protruding abdomen; c) autopsy showing Lung hypoplasia (blue arrow), large bladder (yellow arrow); d,e) Dextrocardia- autopsy showing right-sided heart (green arrow), thymic hyperplasia (orange arrow), removed organs showing right-sided heart (red arrow); f,g) Conjoint twin; f) X-ray picture; arrow-joined skull, thorax and lumbar regions; g) Monochorionic-monoamniotic twin showing two heads, one trunk, a single heart, four upper limbs, and four lower limbs; h) HP image of single umbilical artery in umbilical cord with gross picture in set.

Anthropometric measurements revealed that the majority of cases had normal anthropometric measurements. For HC, CRL, and CHL, the majority had one standard deviation more than normal for gestational age (20%, 30%, 21.4% respectively), while in CC and FL, many had one standard deviation less than normal (16.6%, 22.2% respectively) [Table/Fig-9].

Examination of the placenta and umbilical cord was done in all cases. Placental gross and microscopic examination was within the normal range in all cases. Umbilical cord examination mostly revealed normal findings, with isolated Single Umbilical Artery (SUA) in one case and associated with Edward syndrome in another case.

Cytogenetic testing was possible in 9 (47.4%) cases. 5 (26.3%) cases had a normal karyotype, and one case each of Trisomy 13, Trisomy 18, Trisomy 21, and Monosomy X (FISH) [Table/Fig-10]. Out of these cases, in seven cases, the findings correlated with those of autopsy diagnosis, while in two cases, cytogenetics differed from autopsy findings.

Antenatal USG findings were compared with autopsy findings, and out of 19 cases, in 2 (10.5%) cases, USG findings were proven to be discordant. In 11 cases (57.9%), autopsy added additional findings. In one case, USG diagnosis was omphalocele, but the autopsy revealed anencephaly with encephalocele. In the second case, USG showed anencephaly, whereas the autopsy revealed meningomyelocele. In all cases, the cause of death was found to be congenital anomalies not compatible with life [Table/Fig-11].

## DISCUSSION

The aim of foetal postmortem evaluation is to determine the cause of death, which can provide information about recurrences in future pregnancies, helping in prevention and management, as well as data documentation. Additionally, it serves as a quality control check for antenatal Ultrasound (USG) and a determinant for necessary genetic analysis. The present study was conducted to evaluate the role of each of these factors and to correlate the findings of perinatal autopsy with those of USG and genetic testing. In this prospective study, 19 perinatal autopsies were carried out in the Department of Pathology at SCB Medical College, Cuttack, over a period of two years. The cohort included malformed foetuses, aborted foetuses, and neonates not compatible with life.

Gestational Age (GA) ranged from 19 to 35 weeks, with the majority of cases (36.8%) falling between 20-25 weeks of gestation. Studies by Patel ZM et al., Rasmussen SA et al., and Parmar A et al., showed that preterm babies had four times higher incidence of congenital malformations compared to term babies [10-12]. The present study showed a male preponderance with eight cases, which was similar to the findings of Sridhar K, Taksande A et al., and Sarkar

| GA | Cases | HC | | | CRL | | | CHL | | | CC | | | FL | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | ≤1SD | N | ≥1SD | ≤1SD | N | ≥1SD | ≤1SD | N | ≥1SD | ≤1SD | N | ≥1SD | ≤1SD | N | ≥1SD |
| <19 | 02*,** | 01 | - | - | 01 | - | - | 01 | - | - | 01 | - | 01 | - | 02 | - |
| 20-25 | 07*,*** | 01 | 04 | 01 | 02 | 03 | 01 | 02 | 03 | - | 03 | 03 | 01 | 02 | 05 | - |
| 26-30 | 06* | - | 05 | - | - | 03 | 02 | - | 03 | 02 | 01 | 04 | 01 | - | 06 | - |
| 31-35 | 03 | - | 01 | 02 | - | - | 03 | 02 | - | 01 | 02 | 01 | - | 02 | 01 | - |
| Total cases | 18** | 02 | 10 | 03 | 03 | 06 | 06 | 05 | 06 | 03 | 07 | 08 | 03 | 04 | 14 | - |
| | | 15 | | | 15 | | | 14*** | | | 18 | | | 18 | | |

**[Table/Fig-9]:** Foetal anthropometric measurement.
GA: Gestational age in weeks; HC: Head circumference; CHL: Crown heel length; CRL: Crown rump length; CC: Chest circumference; FL: Foot length; *HC, CHL, CRL not recorded in anencephalic fetus (3 cases); **All anthropometric parameters of conjoint twin not included; ***CHL not calculated in Down syndrome case due to presence of only foot

| Results | No. of cases | % |
|---|---|---|
| Normal | 5 | 26.3 |
| Abnormal | 4 | 21 |
| Trisomy 13 | 1 | 5.3 |
| Trisomy 21 | 1 | 5.3 |
| Trisomy 18 | 1 | 5.3 |
| Monosomy X | 1 | 5.3 |
| Test failure | 4 | 21 |
| Test could not be done | 6 | 31.5 |

**[Table/Fig-10]:** Cytogenetic findings.

| Diagnostic method | Results | No. of cases | Percentage (%) |
|---|---|---|---|
| Autopsy proven cases vs total cases | Diagnosis confirmed on autopsy | 17 | 89.5 |
| | Not confirmed on autopsy | 2 (Trisomy 21, monosomy X | 10.5 |
| | Total | 19 | 100 |
| Antenatal USG vs autopsy findings | Similar findings | 06 | 31.6 |
| | Autopsy added additional information | 11 | 57.9 |
| | Autopsy differed from USG | 02 | 10.5 |
| Autopsy findings vs cytogenetic findings | Similar findings | 07 | 36.8 |
| | Cytogenetics differed from autopsy results | 02 | 10.5 |

**[Table/Fig-11]:** Diagnostic method-wise analysis of results.

S et al., [13-15]. In the present study, the incidence of congenital malformed foetuses was higher in the maternal age group of 21-40 years (16 cases=84.2%), similar to the study by Naik SS et al., [16]. However, there are conflicting results from prior studies, as some have reported a higher prevalence of congenital anomalies in women aged over 35 years [17-21].

An association between the severity of anaemia and foetal malformation was observed by Shi H et al., They reported that 74% of mothers with malformed babies had anaemia. In their research, they found the following Odds Ratios (OR) for the association between anaemia severity and foetal malformation: mild: aOR, 1.15 (95% CI, 1.14-1.17); moderate: aOR, 1.19 (95% CI, 1.16-1.21); severe: aOR, 1.62 (95% CI, 1.52-1.73) [22]. The study also noted that the head and neck were the most common systems showing external defects, which aligned with the findings of Andola US et al., in 2012 [23]. In the present study, the musculoskeletal system accounted for 5 cases (26.3%) of congenital malformations, which was higher than the percentage reported by Tomatir AG et al., (14.2%) [24]. The results varied among different authors, which could be attributed to demographic factors or the smaller number of participants [16, 25-27].

The study did not include anthropometric parameters of the conjoined twins. Measurements such as HC, CHL, and CRL were not recorded in anencephalic foetuses (3 cases) due to the absence of the calvaria. Additionally, CHL was not calculated in one case of DS as only the foot was present without other parts of the extremity. The study showed minor variations in anthropometric measurements, mostly within one standard deviation. This suggests that there is no relationship between foetal measurements and the type of congenital anomalies. Previous studies on the involvement of different organ systems are presented in [Table/Fig-12] [16,24,25,27].

| Authors | Place and year of the study | Sample size | CNS | Musculoskeletal | Genitourinary |
|---|---|---|---|---|---|
| Singh A and Gupta RK, [25] | Jammu (2009) | 140 | 20.5 | 30.6 | 4.7 |
| Boyd PA et al., [27] | United Kingdom (2004) | 309 | 21 | 5 | 3 |
| Tomatir AG et al., [24] | Turkey (2009) | 183 | 31.1 | 33 | 2.3 |
| Naik SS et al., [16] | Andhra Pradesh (2017) | 46 | 33.33 | 16.66 | 8.33 |
| Present study | | 19 | 36.8 | 57.8 | 26.3 |

**[Table/Fig-12]:** Comparison of system involvement with other authors [16,24,25,27].

In seven cases, autopsy findings were in concordance with USG, and all of them underwent karyotype analysis. Abnormalities were noted in two cases (DS, TS), which helped predict the cause of death and confirm the diagnosis. In the remaining cases, the diagnosis was based on both autopsy findings and observed abnormalities in USG (89.4%). This percentage was comparable to the findings of Vogt C et al., who reported complete agreement between prenatal ultrasound and postmortem findings in 84% (384/455) [28]. However, in two cases, autopsy revealed that the initial USG findings were incorrect (1-anencephaly with encephalocele, 2-meningomyelocele). Both cases underwent genetic analysis, which showed normal results or test failure. As a result, autopsy played a pivotal role in diagnosing these two cases and predicting the cause of death.

In 10 cases, autopsy revealed additional findings such as ES, TD, PS, HS, and conjoined twin. Genetic testing was performed in four of these cases. Two cases were found to have abnormalities (ES and PS), while the tests failed to yield any result in the other two cases. Therefore, these cases were diagnosed again based on autopsy findings. The correct autopsy diagnosis was obtained in 17 out of 19 cases. In two cases, autopsy failed to provide a confirmed diagnosis (Trisomy 21 and Monosomy X) because not all the findings necessary for a confirmed diagnosis were detected. These cases were later diagnosed using cytogenetic study. Aggarwal S et al., study demonstrated that dysmorphological examination confirmed a definite genetic diagnosis in sixteen cases, histopathology in six cases, and karyotyping, biochemical testing, and exome sequencing in two cases each [29]. This analysis also reveals that while USG plays an important role, it may fail to provide a correct final diagnosis. Perinatal autopsy has a major advantage in documenting all abnormal findings, indicating the necessity and type of genetic analysis required. It is important to note that a combination of USG, autopsy, and genetic evaluation is essential for obtaining optimum results in the approach to congenitally malformed and deceased foetuses, as well as for managing such cases and providing parental counselling for future pregnancies.

## Limitation(s)

Although there is a small number of cases, the incidence of congenital malformations causing perinatal deaths, especially the syndromes encountered here, has a very low prevalence rate.

## CONCLUSION(S)

Foetal and perinatal autopsies are an essential part of the clinical management for families experiencing the loss of a foetus or newborn. The present study observed a correlation between USG and autopsy in 31.6% of cases; however, additional findings were observed in 57.9% of cases. In 10.5% of cases, USG was found to be incorrect. A confirmed diagnosis was made by autopsy alone in 89.5% of cases, but in 10.5% of cases, the diagnosis was confirmed after cytogenetic study. When combined with genetic testing, the results are more reliable and confirmed. Although USG is easy and preferred, and can provisionally indicate anomalies, autopsy can contribute to and modify the findings, aiding in the documentation of cases. Therefore, a comprehensive approach that includes dysmorphological evaluation, imaging studies, and genetic testing, as indicated, has a high diagnostic performance and is vital for establishing the final diagnosis.

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