Abstract: Background: It has been seen that there is progressive loss of Purkinje cells in various genetic conditions like ataxia telangiectasia and Niemann Pick disease type C. So, this study was carried out to analyse the histogenesis of human fetal cerebellar cortex. Methods: The study was conducted in the 30 aborted foetuses. The development of various layers of the cerebellar cortex is studied. All the layers of cerebellum were measured using image analyzer software-image pro premier 9.1. Data analysis was done by regular Statistical method. Results: The thickness of the 3 layers shows a gradual increase that indicates the developmental aspect of the 3 layers of the Embryo. The Purkinje cells appeared during the 4th month of embryonic period. Conclusion: The present study will facilitates in the identification of various pathological lesion of cerebellum which may develop in intrauterine life of fetus.

Key Words: Cerebellum, Purkinje cell, Aborted Fetus, Gestational Age, Niemann Pick disease type C

Introduction:
The cerebellum is a part of the hindbrain located above the medulla in the posterior cranial fossa. Cerebellum is the part in the brain that first start differentiating but it is the last one to complete its maturation. As, its development range over a longer period we can study age related changes in it very well. (1) There are two parts of cerebellum an outer cortex of gray matter and an inner central core of white matter. There are three well-defined layers of cerebellar cortex from inside to outside. These are (2):
1. Granular layer: It consist of densely packed granule cells, along with Golgi cells.
2. Purkinje layer: it consist of a single strip of cell bodies of Purkinje cells and Bergmann glial cells.
3. Molecular layer: It contains the flattened dendritic trees of Purkinje cells, along with the T shaped parallel fibres of granule cells penetrating the Purkinje cell dendritic trees at right angles. It contains two types of inhibitory cells the stellate cell and basket cell. They synapses onto Purkinje cell dendrites. Basket cells synapse on the Purkinje cell axon initial segment and stellate cells onto its dendrites.

In fetus, an additional layer called, external granular layer is also seen which is the precursor of Purkinje cell layer and internal granular cell layer. (3) The Purkinje cells finally project onto the deep cerebellar nuclei. They are the lune output cells of the cerebellar cortex.

Obersteiner defined the external granular layer more precisely and it was thus called Obersteiner’s layer. He differentiated it into two layers. He found that the cells in the superficial layer form the basal membrane and they either transform into glia or become atrophied, while cells of the internal layer move into the molecular layer and travel through this into the inner granular layer. (4) So, the aim of this study was to analyse the histogenesis of human fetal cerebellar cortex and to study the origin and development of Purkinje cells.

Materials and Methods
The study was conducted on 30 aborted human fetuses. Gestational age of fetuses, was found out by assessing their Crown- rump and Crown- heel length. Total number of fetuses were divided into 3 gestational age groups-
Group 1: Gestational age is <12 weeks (10 fetuses)
Group 2: Gestational age 13-24 weeks (10 fetuses)
Group 3: Gestational age > 24 weeks (10 fetuses)

Institutional Ethics Committee clearance was taken before the beginning of the study. Foetal cerebellum specimens were dissected and exposed to routine histological processing. After processing the paraffin tissue blocks were made and they were cut in to 5µm thickness and slides were made later they were processed using haematoxylin and eosin stain.
stained with hematoxylin and eosin. Then the mounted slides were visualized under low power and high power objectives of binocular light microscope for various histological developmental features of the cerebellum like appearance of external granular layer and Purkinje cells.

The width of all the 3 layers was measured in the histological features of the cerebellum using image analyzer software-image pro premier 9.1. The mean was calculated using the regular statistical methods.

Results
In our study we found that Purkinje cells appeared during the 16th week of embryonic period. We also noticed in our study a gradual increase in the thickness of the 3 layers that indicates the developmental aspect of the 3 layers of the Embryo. We also observed the presence of external granular layer in 1st trimester. Histological sections of each trimester showing each layer is shown in Figures 1, 2 and 3. Appearance of Purkinje cells is shown in Figure 2.

![Fig 1 Histological section of cerebellum showing each layer in 1st trimester. EGL- External granular layer, IGL- Internal granular layer, ML- Molecular layer](image)

![Fig 2 Histological section of cerebellum showing each layer in 2nd trimester. EGL- External granular layer, ML- Molecular layer, PL- Purkinje layer, IGL- Internal granular layer.](image)

![Fig 3 Histological section of cerebellum showing each layer in 3rd trimester. ML- Molecular layer, PL- Purkinje layer, IGL- Internal granular layer.](image)

The mean thickness of each layer in all trimester is shown in Table 1.

<table>
<thead>
<tr>
<th>Trimester's</th>
<th>Molecular layer (Mean)</th>
<th>Purkinje layer (Mean)</th>
<th>Granular layer (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>182.603mm</td>
<td>Not Formed</td>
<td>899.794mm</td>
</tr>
<tr>
<td>2</td>
<td>210.529mm</td>
<td>17.87mm</td>
<td>983.688mm</td>
</tr>
<tr>
<td>3</td>
<td>263.287mm</td>
<td>34.549mm</td>
<td>1048.894mm</td>
</tr>
</tbody>
</table>

Discussion
During the embryonic life the sub ventricular zone forms at the edge of the rhombic lip. Later its cells migrate over the superficial surface of the whole cerebellum and they proliferate as they travel, to form a temporary embryonic structure which consists of extremely abundant small cells. This layer is known as external granular layer. Neurons in the external granular layer continue their cell division for a long time than any other neurons in other parts of the brain. This can leads to tumour development. (5)

D.Asha latha et al, found that the external granular layer appears as a single layer at 16 weeks and later it became bilayer at 20 weeks and tri-layered at 26 weeks of gestation. She also stated that external granular layer is the originator of the molecular, Purkinje and internal granular layers of the cerebellum. The EGL gradually decreases as the age of foetus increases and this descent is rapid and is completely lost after postnatal 9th month. They also found that the Purkinje cells are appreciable and multi cellular arranged in the form of clusters beneath the External granular layer at around 20 weeks. (5)

Rakic P and Sidman RL found that the Purkinje cells develop by 13 weeks. (6) Veni SK et al found that the Purkinje cells differentiate during 12th -16th week which was observed in the current study also. They also found that a thin external granular layer is seen in peripheral zone at 13 weeks in our study also we found the appearance of external granular layer in 1st trimester. (7)

According to Schienker the cells of external granular layer of cerebellum can remain postnatally and form the basis for the origin of medulloblastoma. (8) Christos et al. in his study of human cerebellum also observed that medulloblastoma, which is seen most commonly in malignant central nervous system tumours in children are traceable to cerebellar embryogenesis. (9) This study will be helpful in diagnosing these conditions.

Conclusion
The thickness of the different layers were assessed in the current study. It was observed in the current study that the 3 layers shows a gradual development, wherein its thickness increases as progressive development takes place. The Purkinje cells appeared during the 16th week of embryonic period. The present study offers an insight of the microscopic alterations during the embryogenesis of cerebellum it may aid in identifying the pathological lesion of cerebellum during the intrauterine life of fetus.

References