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Anatomical studies on the pancreas of sheep (*Ovis aries*)

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Abstract

Pancreas was pale pink coloured, lobulated gland located along the median plane in the abdominal cavity. In group I, Islets of Langerhans appeared at 47 days of gestation. In group II, at 77 days of gestation, different types of cells viz., alpha, beta and delta cells were found intermingled with each other. In group III, at 118 days of gestation, the mean length and width of the islets were increased. In group IV, Islets of different sizes and shapes appeared as pale cluster of cells. The mean length and width of the islets decreased with age. The cells of islets of Langerhans showed mild PAS, Millon's reaction, weak acid phosphatase and strong alkaline phosphatase, alpha naphthyl acetate esterase reactions in all the age groups studied.

Keywords: Gross morphology, Histochemistry, Histology, Pancreas, Sheep

Introduction

Pancreas of small ruminants is a major accessory organ and has an important role in digestion. Endocrine part of pancreas is Islets of Langerhans function to maintain glucose homeostasis (Mahesh *et al.* 2017) ^[9].

Knowledge of development of pancreas is essential to understand congenital pancreatic abnormalities arising from abnormal histogenesis and morphogenesis occurring during critical period of organization of the organ during prenatal life (Tadokoro *et al.* 2011) ^[18].

Nowadays, the biology of the pancreas is being studied intensely, with hope of finding better treatments for devastating pancreatic diseases, such as diabetes mellitus, pancreatitis, and pancreatic adenocarcinoma. In particular, advancements in stem cell technology have recently sparked optimism that diabetes could be cured by harvesting stem cells for therapeutic use. This has led to heightened interest in understanding embryonic development of the pancreas (Shih *et al.* 2013) ^[19].

Materials and Methods

The present work was conducted at the Department of Veterinary Anatomy, Veterinary College and Research Institute, Namakkal. The approximate age in prenatal age groups was calculated by the formula given by Richardson, 1980 (Noakes *et al.*, 2009) ^[12].

$$X = 2.1(Y + 17)$$

'X' is the developmental age of fetus in days and 'Y' is crown rump length in centimeters. In postnatal age groups, the age was ascertained based on the eruption of teeth as described by Dyce *et al.* (1996) ^[4].

Samples were collected from six different age groups with six animals in each group.

The groups are classified as follows

Group I (1-50 days of gestation)	Group II (51-100 days of gestation)	Group III (101-150 days of gestation)	Birth to 3 months (Prepubertal)	7-9 months (Pubertal)	Group VI (2 years and above, adult)
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Results and Discussion

Pancreas in sheep fetus upto fifty days of gestation was pale pink coloured, lobated gland. It was located along the median plane, in the abdominal cavity extending over the mesentry. This was in uniformity with observations of Singh and Sethi (2012) ^[13] in buffalo fetuses Al - Saffar *et al.* (2014) ^[1] in rabbits and Divya *et al.* (2017) ^[3] in buffalo fetuses.

Part of the gland extended into the loop of duodenum, rumen and abomasum. Posteriorly it was related to the right and left adrenal glands was similar to findings of Singh *et al.* (2017a) ^[14] in the pancreas of fetal goat.

Pancreas in sheep fetus from fifty-one to hundred days of gestation was creamish pink in colour, located dorsally in the abdominal cavity within the loop of the duodenum.

Most of it was located towards left of the median plane, while a part of the gland crossed the median plane, entered the right half and extended up to the antero-medial extremity of the right kidney. It was related anteriorly to the rumen and omasum, posteriorly to the left adrenal gland and colon (Fig 1). This was as observed by Singh *et al.* (2017a) ^[14] pancreas of fetal goat.

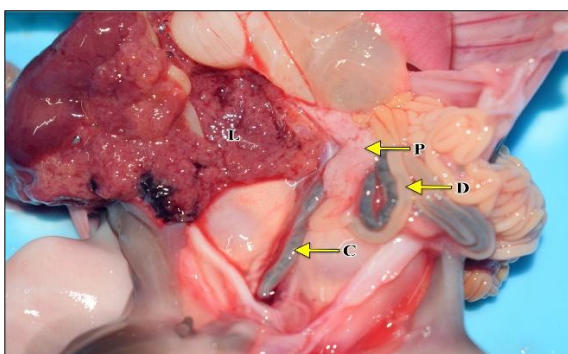


Fig 1: Photograph showing pancreas in sheep fetus at 77 days of gestation

P-Pancreas L-Liver C-Colon D-Duodenum

Pancreas in sheep fetus from hundred and one days to full term gestation was located dorsally in the left side of the abdominal cavity. It was creamish pink in colour located within the loop of duodenum was as reported by Iniyah *et al.* (2019) reported that the pancreas in large white Yorkshire.

Pancreas was related anteriorly to the omasum, abomasum and antero-dorsally extended up to the muscular rim of the diaphragm. Posteriorly this gland was related to the colon and partly to the anterior extremity of the left adrenal gland (Fig 2). T was in accordance with findings of Divya *et al.* (2017) ^[3] in pancreas of buffalo fetuses and Singh *et al.* (2017a) ^[14] in fetal goat.

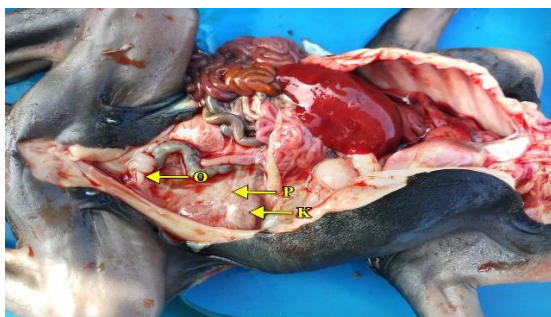


Fig 2: Photograph showing pancreas in sheep fetus at 110 days of gestation

P-Pancreas, O-Ovary, K-Kidney

Pancreas in sheep was creamish white in colour, located within the duodenal loop as reported by Singh *et al.* (2017a) ^[14] in goat fetuses.

It was related anteriorly to the lobes of the liver, laterally to the right kidney and right adrenal glands and ventrally to the large intestines. Small portion of the gland was extended over the rumen (Fig 3) as studied by Divya *et al.* (2017) ^[3] in pancreas of buffalo fetuses and Singh *et al.* (2017a) ^[14] in fetal goat.

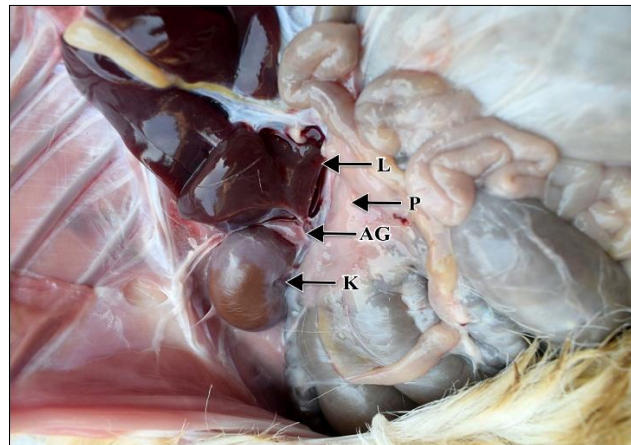


Fig 3: Photograph showing pancreas in 2months old sheep
P-Pancreas, AG-Adrenal Gland, K-Kidney, L-Liver

In the pancreas of sheep at 47 days of gestation, encapsulated islets of Langerhans which appeared as the ovoid aggregation of cells with clear peripheral space were observed in between the acini (Fig 4). These observations were similar to the findings of Gupta *et al.* (2002) ^[15] in human fetuses ranging from 12- 18 weeks of gestation. On contrary, Sreeranjini *et al.* (2015) ^[16] found that at 69 days of age, several developing islets of Langerhans were seen as groups of cells of varying sizes scattered at the periphery of ductules. Hisaoka *et al.* (1992) ^[7] found primordium for pancreatic islets at 12 days of gestation and Singh and Sethi (2012) ^[13] noticed the appearance of endocrine cells at 125 days of gestation in buffalo fetuses.

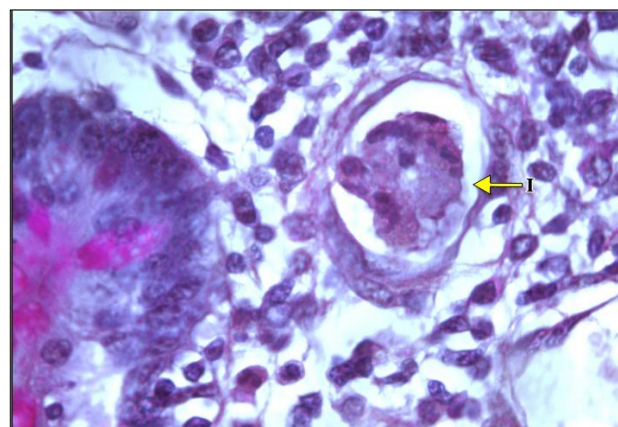


Fig 4: Photograph showing pancreas in sheep fetus at 47 days of gestation

I-Islets of Langerhans, PAS X 1000

At 77 days of gestation, numerous encapsulated islets of different sizes and shapes were observed (Fig 5). The length and width of the islets of Langerhans increased when compared to group I (Table 1).

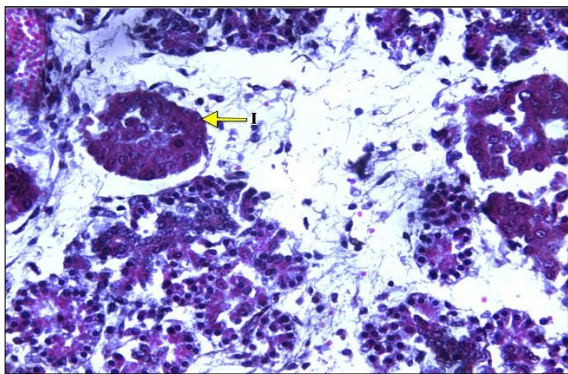


Fig 5: Photograph showing pancreas in sheep fetus at 77 days of gestation
I- Islets of Langerhans, H&E X 400

Mean (\pm S.E.) values of histometrical parameters of islets of Langerhans in pancreas of pre and postnatal age groups in sheep

Group	Mean length of islet of Langerhans(μ m)	Mean width of islet of Langerhans(μ m)
I	50.5235 \pm 0.96	43.41 \pm 2.05
II	89.38 \pm 11.07	50.52 \pm 4.65
III	91.21 \pm 12.05	63.12 \pm 7.23
IV	92.69 \pm 7.69	70.90 \pm 5.20
V	72.09 \pm 5.08	47.22 \pm 3.56
VI	71.97 \pm 5.33	46.12 \pm 5.29

Different types of cells viz., alpha, beta and delta cells were found intermingled with each other. The cells were eosinophilic with vesicular nucleus. When stained with Maldanado stain, alpha cells appeared pink, beta cells were violet and delta cells were light blue in colour (Fig 6). Some islets contained blood filled spaces within them. These observations were in uniformity with the reports of Sreeranjini *et al.* (2015) [16] goat fetuses.

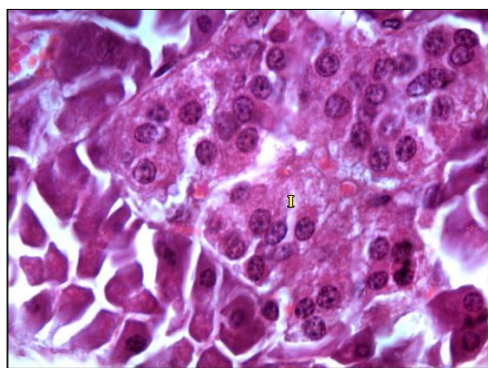


Fig 6: Photomicrograph showing pancreas in 3 year old sheep
I- Islets of Langerhans, H&E X 1000

At 118 days of gestation, the mean length and width of the islets were increased compared to group II while rest of the histological features were identical (Table 1). In pancreas of sheep upto three months of age, islets of Langerhans of different sizes and shapes appeared as pale cluster of cells compared to cells of exocrine part of the gland which was in accordance with the findings of Tsuchitani *et al.* (2016) [17] in pancreas of rat and Mahesh *et al.* (2017) [9] in Deccani sheep and Badri goat. Within the islets of Langerhans alpha, beta and delta cells were found intermingled with each other. These observations were in disagreement with the reports of a

Meshram *et al.* (2001) [11] in pancreas of goat, Tsuchitani *et al.* (2016) [17] in pancreas of rat, Mahesh *et al.* (2017) [9] in the pancreas of Deccani sheep and Badri goat who opined that alpha cells were located towards the periphery of the islet, beta cells were located at the center of islet and delta cells were located at the periphery of the islet intermingled with alpha cells.

The islets were not encapsulated and delineation between the islets and exocrine portion of pancreas was not distinct (Fig 7). The mean length and width of the islets of Langerhans were increased when compared to prenatal groups, which was in accordance with Mansouri *et al.* (2006) [10] in sheep pancreas.

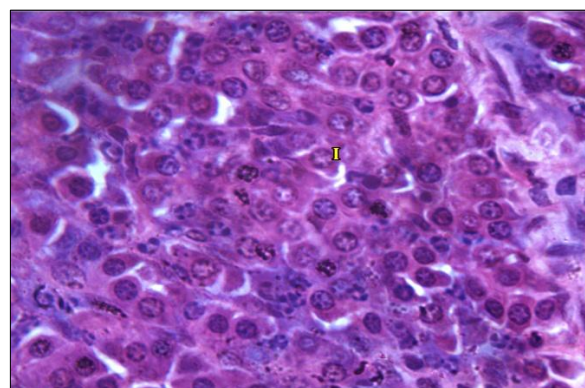


Fig 7 Photomicrograph showing pancreas in 2 months old sheep
I- Islets of Langerhans, H&E X 1000

In pancreas of sheep between seven and nine months of age the mean length and width of the islets of Langerhans decreased compared to group IV (Table 1). In contrast, Mansouri *et al.* (2006) [10] reported an increase in the diameter of islets in pancreas of sheep from new born to sexually mature groups.

In pancreas of sheep above two years of age, pancreas consisted of round to irregular shaped small, medium and large islets of Langerhans distributed throughout the parenchyma of the gland which was same as the reports of Meshram *et al.* (2001) [11] in pancreas of goat.

The islets were separated by a very narrow space from the surrounding exocrine portion of the gland. They were richly supplied by blood vessels (Fig 8). These observations were in contrary with the findings of Balasundaram (2018) [2] who reported that in pancreas of goat the islets were distributed along the periphery of pancreatic lobes.

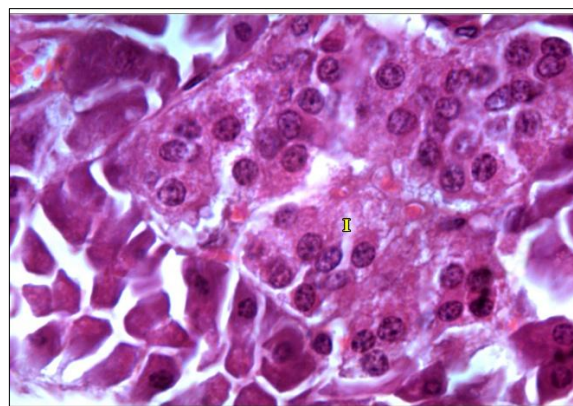


Fig 8: Photomicrograph showing pancreas in 3 years old sheep
I- Islets of Langerhans, H&E X 1000

The mean length and width of the islets of Langerhans decreased compared to group V. This observation was same as noticed by Mansouri *et al.* (2006)^[10] in sheep pancreas. In the pancreas of sheep, the cells of islets of Langerhans showed mild PAS positive reaction in all the age groups studied (Fig 9) which was in agreement with observations of Singh *et al.* (2017b)^[15] in fetal goat pancreas.

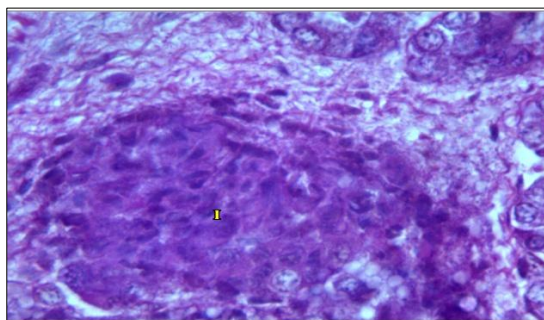


Fig 9: Photomicrograph showing PAS reaction in pancreas of sheep fetus at 94 days of gestation
I- Islets of Langerhans, PAS X 1000

In all the age groups studied, the capsule, cells of the gland and connective tissue fibres in the pancreas were mildly positive for Millon's reaction indicating the presence of tyrosine. (Fig 10)

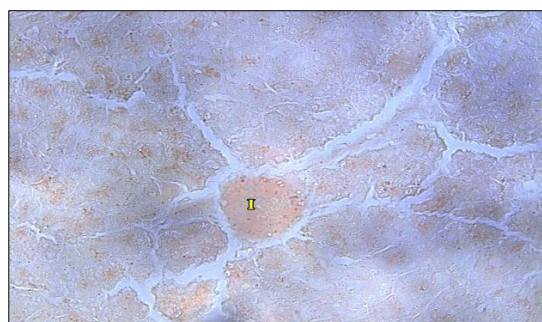


Fig 10: Photomicrograph showing localization of tyrosine in 7 months old sheep
I- Islets of Langerhans, Millon's reaction X 400

In the pancreas of sheep, Islets of the Langerhans in the pre and postnatal age groups showed weak reaction for the presence of acid phosphatase (Fig 11) which was in accordance with findings of Hellerstrom (1963)^[6] in duck pancreas. Whereas, Singh *et al.* (2017b)^[15] reported that in fetal goat pancreas islet cells, acid phosphatase activity was not observed.

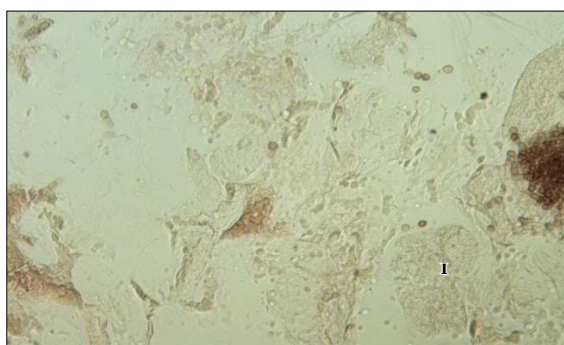


Fig 11: Photomicrograph showing acid phosphatase reaction in 3 months old sheep
I- Islets of Langerhans, Azo Dye coupling Method X 400

Islets of the pancreas showed strong positive reaction for the presence of alkaline phosphatase in all the age groups studied which was as reported by Hellerstrom (1963)^[6] in duck. Whereas, Singh *et al.* (2017b)^[15] reported mild positive reaction for alkaline phosphate in late prenatal period in fetal goat pancreas islet cells. Pancreas showed strong positive reaction for alpha naphthyl acetate esterase activity in pre and postnatal groups studied. (Fig 12).

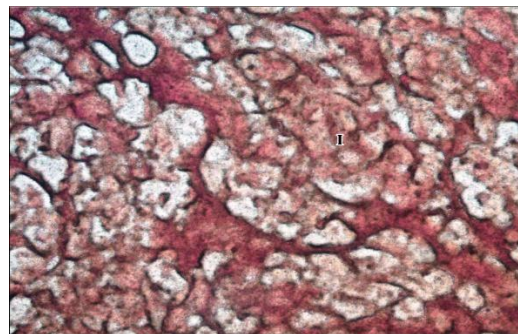


Fig 12: Photomicrograph showing alpha naphthyl acetate reaction in sheep fetus at 128 days gestation
I- Islets of Langerhans, Alpha naphthyl acetate method Method X 400

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