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COMPARATIVE STUDY OF THE PANCREAS HISTOLOGICAL AND HISTOCHEMICAL IN BARN OWL (TYTO ALBA) AND BLACK FRANCOLIN (FRANCOLINUS)

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Abstract: The present study aimed at the pancreas of two bird species differs in their classification, habitat, nutrition, and activity. The birds were omnivorous Black Francolin(francolinus) and carnivorous Barn Owl (Tyto alba). Histologically the pancreas of these birds was covered by a thin layer of connective tissue capsule formed from reticular, elastic, and few collagenous fibers, this capsule in barn owl appear thicker than that found in black francolin birds. The parenchyma of pancreas in both birds were composed mainly from exocrine portion represented by serous acini and endocrine portion which distributed among acini which represented by islets of Langerhans. The endocrine gland of the pancreas showed that the islets of Langerhans, have two types of cells, the first one is alpha cells shown as a spindle in shape with oval nuclei, located periphery of the islets of Langerhans. The second type was beta cells show as rows or irregular polygonal cells with spherical nuclei that is slightly larger than the nuclei of alpha cells and found in the center of islets. These cells were located in the periphery of the island of Langerhans in both birds. Histochemical results reveal that the pancreatic cells, whether the acinar cells or islets cells are moderately positive in both species of birds.

Keywords: -

Introduction

Birds are consider as one of bigger classes in kingdom of Animalia including (9990) species which distributed in twenty orders, many countries legislated some laws to protect birds from hunting in Breeding season to prevent it from extinction (Abu- al Hubb ,1994) and(Grimmett , Inskipp, 2011).

The digestive system is a long hollow tube or tract that has many functions such as digestion and absorption of nutrients that are necessary for organisms, the system begins at the beak and finishes at the vent in sequential order it is composed of a mouth, esophagus, crop, proventriculus, ventriculus (gizzard), intestine, caeca, rectum and cloaca. However, associated with the digestive tract, the accessory digestive organs which include the liver, pancreas, and gall bladder deliver their secretory

products to the small intestine by excretory ducts, which are the common bile duct from the liver and the main pancreatic duct from the pancreas that join in the duodenal loop to form a single duct common to both organs (Eroschenko, 2008), (Denbow, 2015) and (Ali and Hussein, 2020).

The pancreas is a soft, elongated, and vital lobulated organ in the digestive system, located on the right side of the abdominal cavity in all birds (Al-Sharoot, 2016). However, it is a mixed exocrine-endocrine gland that produces digestive enzymes and hormones, the exocrine portion of the pancreas is a compound tubuloacinar gland that produces digestive enzymes that are routed to the intestine by a branched ductal network, whereas, the hormones synthesized by the endocrine cells of the pancreatic islets such as insulin, glucagon, somatostatin, and pancreatic polypeptide (PP) that is released into the blood circulation (Frappier, 2007, Hafez etal., 2015 and Ali ; Hussein, 2020). The current study aimed to comparative study of the pancreas histological and histochemical finding of the above birds

Methods

Ten birds were used to conduct the current study. They were distributed into two groups each comprising five birds of the selected male species. The two different species used are Barn Owl (Tyto alba) and Black francolin (francolinus) as a carnivores and omnivore birds, respectively. These birds were collected from common markets at Al-Samawa City. The birds were bought from the local suppliers at these markets.

Birds were weighed before euthanasia. The birds were euthanized prior to their dissection with an intramuscular injection of sodium pentobarbitone (80 mg/kg) (Mitchell and Smith, 1991). After that, it was dissected by fixing them on a dissecting board. A mid-line incision was made in the abdominal wall of each bird to view the abdominal viscera. This organ was identified, the pancreas of the dissected birds was well described. The organ then after were washed by normal saline to remove blood or any other adhering debris. The contents of the pancreas were emptied by gentle pressure on each of them, and then washed by normal saline again.

For histological study, half of the specimens (pancreas) from each dissected birds were collected and fixed in Bouin's solution and the other half were fixed in 10% neutral buffered formalin. After well fixation the specimens were dehydrated by passing them through a series of ascending ethanol alcohol each for two hours (70%, 80%, 90% , 95% and 100%) and then specimens were cleared in xylene for one hour after that embedded in paraffin wax and then the blocks were sectioned at 6µm thickness and stained with either one of the following stains: Mayer's Hematoxylin and Eosin routine stain for general features identification and Masson trichrome stain for the staining of the collagenous and smooth muscle fibers (Bancroft and Stevens, 2010).

To conduct the histochemical study for used Periodic acid Schiff (PAS) for the neutral mucin of the pancreas.

Result and Discussion

The results of the pancreas show that it is covered in each bird by a connective tissue capsule as in Fig. (1) which consists of collagenous and elastic with fibroblast cells that have a flattened shape and oval nuclei, this capsule in barn owl appears thicker than that found in black francolin bird. This result agrees with Hamodi et al., (2013) who mentioned that the capsule appears to be thicker in common gull than in guinea fowl, (Al-Hathry, 2000) who reported that the capsule of common quail was thick and irregularly dense connective tissue. But disagrees with Al-Tikrity and Al-Samarrae, (1992); Hussein, (2017) who reported that the pancreas in local chicken and local broiler fowl was surrounded by a very thin capsule.

The connective tissue septa extend from the capsule into the parenchyma of the pancreas dividing it into many lobules. These septa were very thin and some of them carried blood vessels distributed all over the pancreas. Blood vessels were varied in their sizes and ganglionic nerve

cells were found in perivascular connective tissue of large blood vessels and characterized by oval or rounded pale nuclei with prominent nucleolus.

The parenchyma of pancreas in both birds were composed mainly from exocrine portion represented by serous acini and endocrine portion which distributed among acini which represented by islets of Langerhans fig. (3).

the exocrine was arranged in the form of serous tubuloacinar glands that occupy a large area of the pancreas and have oval to spherical shapes in barn owls while in francolin birds were more oval than spherically shaped. These results were enhanced by Simsek et al., (2009); Faris, (2012), and Al-Sharoot, (2016) who found that in pigeon, early-hatched goose, and in falcon's genus (Falco Anaumanni) , also agree with (Das et al., 2003; Kadhim et al., 2010) who explained that these shapes were based on the plane of section as documented in other birds. In addition, it is composed mainly of a mutable number of acinar cells that have a pyramidal or tall columnar shape. In both birds as in Fig. (2,4)

The ductal system of the exocrine part of the pancreas in both birds includes the intercalated ducts, intralobular ducts, and interlobular ducts. Intercalated ducts are circular, narrow lumen in almost and are lined with a simple flat to cuboidal epithelium in the both birds but it look more regular in francolin than owl, its average diameters in francolin and in owl (25.83)and (23.57) respectively. The intralobular ducts are scattered through the pancreas parenchyma of the two species of birds without a defined arrangement, these ducts have mostly circular shape and are lined by the same epithelial cell that lined the intercalated ducts, its average diameter in francolin fowl (62,14) and in owl (42.5), and they are surrounded by secretory acini (Fig.3,5). The interlobular ducts have an irregular to oval shape and its average diameter in francolin fowl (217) and in owl (361.66). These ducts are composed of an epithelium that appears simple squamous to cuboidal in francolin fowl and simple flattened epithelium, which lines up inside the ducts and connective tissue surrounding outside the ducts. Furthermore, interlobular ducts have thin muscle layers surrounding the connective tissue in the francolin fowl and are absent in owls (Fig.2,4). Also the blood and lymph vessels, nerves, and excretory ducts run within the connective tissue septa in both birds. The structure of the ducts in the two birds found to be similar to that described in previous studies (Gulmez, 2003; Stornelli et al., 2006; Al-Shaeli, 2010, Sayrafi et al., 2011). Moreover, it was similar partially to Beheiry et al., (2018) who reported that the intercalated duct was lined by squamous to low cuboidal epithelium, the intercalated duct extended as intralobular duct and lined by a single layer of simple cuboidal epithelium, while it is dissimilar in the interlobular ducts which are lined by cuboidal epithelium and characterized by lymphocytic aggregation within the wall of the duct. The result does not match partially with Mutoh et al., (1998) who mentioned that the intercalated duct in fowl gave a changeable description of the duct as it was lined by quite inconspicuous cells that seemed to appear as stellate cells.

The endocrine part of both birds consists of various shapes and sizes of islets which are called Langerhans islets that are scattered in the exocrine part, These islets don't have distinct borders with the exocrine part in both birds (Fig.3,5). In the endocrine part of francolin, the mixed islets that consist of A and B cells are observed and also some islets contain only A cells which are known as alpha islets, and beta islets that contain only B cells, the alpha islets are larger than the beta and are more densely populated A cells are more demonstrated than B cells and the nucleus, in both cell types, are seem similar and is round to oval in shape with a prominent nucleolus. These results agree with many other researchers such as (Al-Shaeli, 2010; Kadhim et al., 2010 and Steiner et al., 2010) who showed that three different types of cells have been observed in birds, primarily studies of the white-crowned sparrow, the mallard, and the chicken (Gallus gallus). As well this corresponds to Das et al., (2003); Simsek and Al-Abay, (2008) who explained that the endocrine pancreas of birds contains three types of cells that release glucagon,

insulin, somatostatin, the interaction between these hormones and pancreatic hormone – plasma metabolite feedback mechanisms is the main regulators of these secretions. According to suggestions by Nishida et al., (1992) on chickens and other birds that feed on grains and seed, they need more enzymatic activity to compensate for their lack of teeth and hydrolytic enzymes in their saliva. On other hand, it disagreement with (Al-Agele and Mohammed, 2012) who showed that there are two types of cells in the endocrine part of the pancreas in golden eagles (Aquila chrysaetos). The microscopic examination shows that the alpha cell is spindle in shape with oval nuclei, located periphery of islets of Langerhans. Beta cells are rows of irregular polygonal cells with spherical nuclei that are slightly larger than the nuclei of alpha cells and found in the center of islets. The islets of both birds are very rich in blood capillaries, which appear more in francolin than in owl.

The histochemical results as in Fig (3,5) reveal that the pancreatic cells, whether the acinar cells or islets cells are moderately positive for PAS reactions in both species of birds but the interaction is stronger contains red granules in francolin than in owl, and the acinar cells than in the islets cells. PAS-positive material is also seen in the lumen of acini. This is similar to Hussein, (2017) who concluded that the PAS-positive material can show in the pancreas of local broiler fowl and common moorhen, similar to (Hamodi et al., 2013) who explained that the carbohydrates appear as pink-color deposits as identified throughout the cellular parenchyma of the pancreas, especially in Guinea fowl.

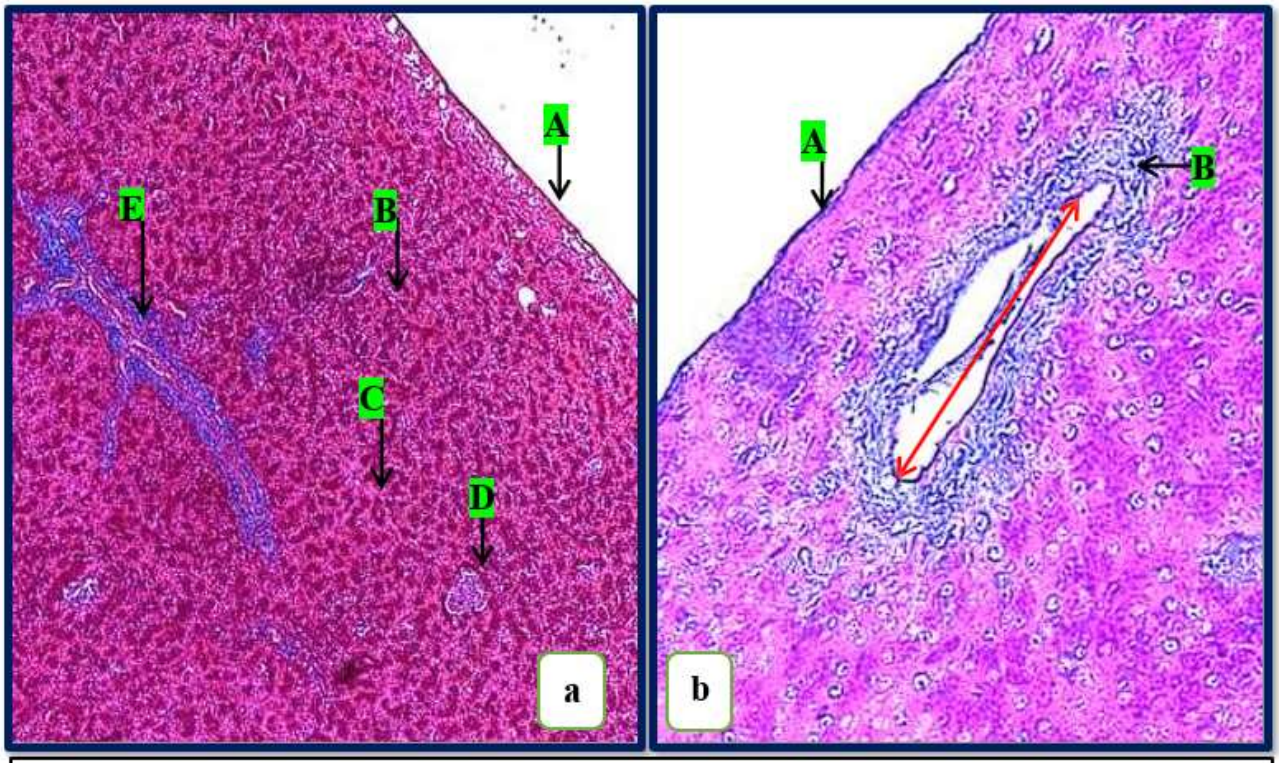

Fig.1. Photographic of the pancreas show the: **A**.capsule, **B.** acinus,**C**.centroacinar cell, **D**. Islet of Langerhans,**E**. Interlobular duct, **F.**connective tissue and double head red arrow (Intralobularduct).

(a) Owl X 40 Masson Trichrome (b) Francolin X400 PAS

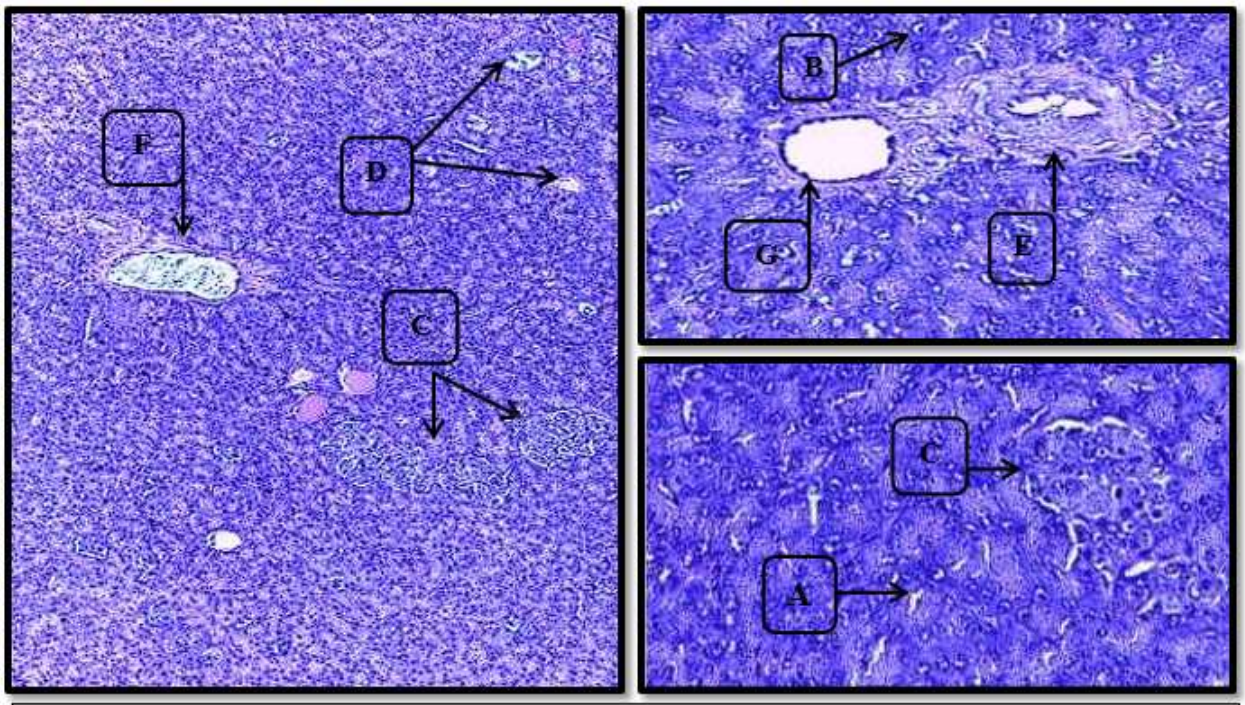

Fig.2. Photographic of the pancreas in owl show the: **A**. acinus, **B**. centroacinar cell, **C**. Islet of Langerhans,**D**. Intercalated duct,**E**. Intralobular duct,**F**. Interlobular duct and**G**. Blood vessel.

(a) X 40 and (b, c) X400 H & E.

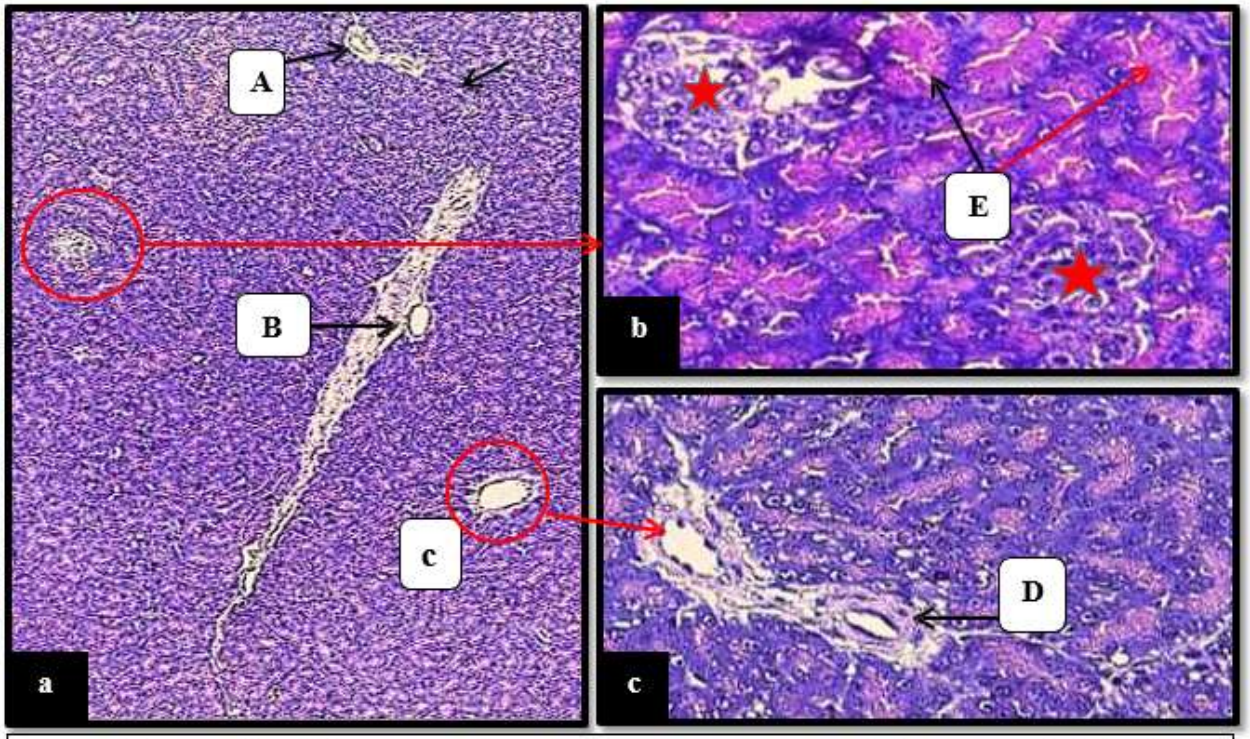

Fig.3. Photographic of the pancreas in owl show the **A**. Intercalated duct, **B**. Interlobular duct, **C**. Intralobular duct,**D** .Wall of blood vessels,**E**.Mucin with acini and star red Islet of Langerhans,

(a) X 40 and (b, c) X400 PAS.

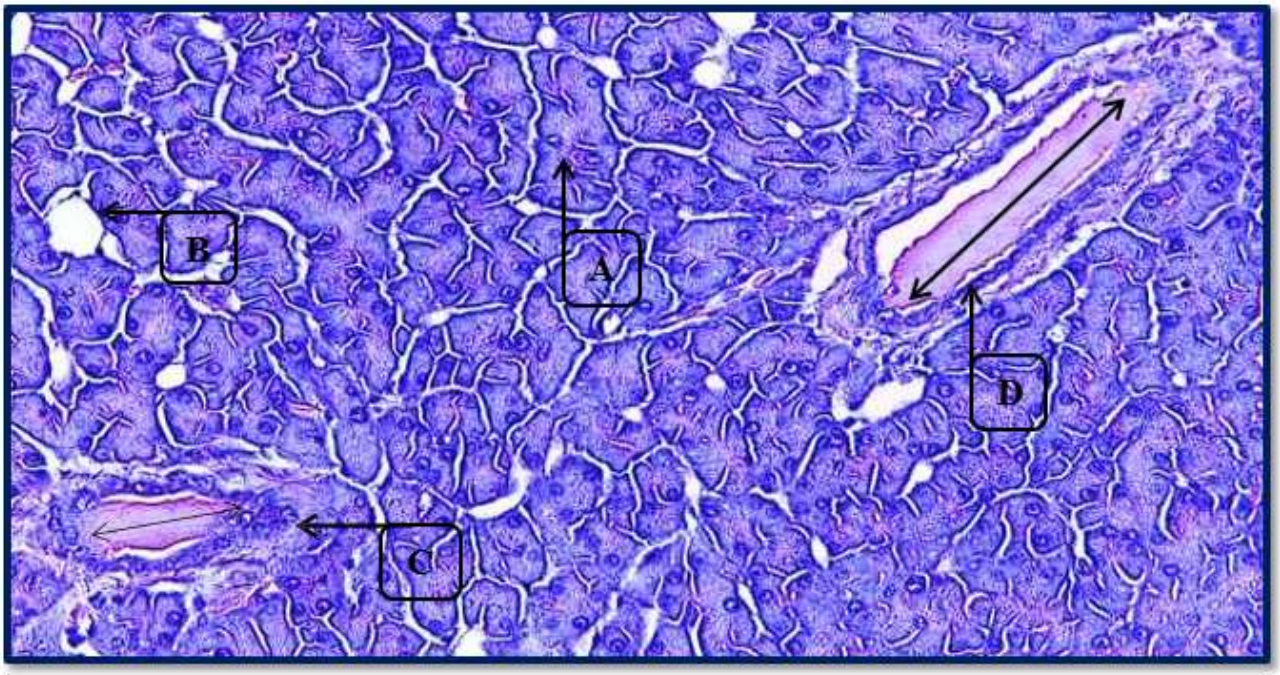

Fig.4. Photographic of the pancreas in francolin show the: **A**. Serous acini, **B**. Centroacinar, **C**. Cuboidal epi., **D**. Secretion of duct and double head black arrow (Intralobular duct).

X400 H & E.

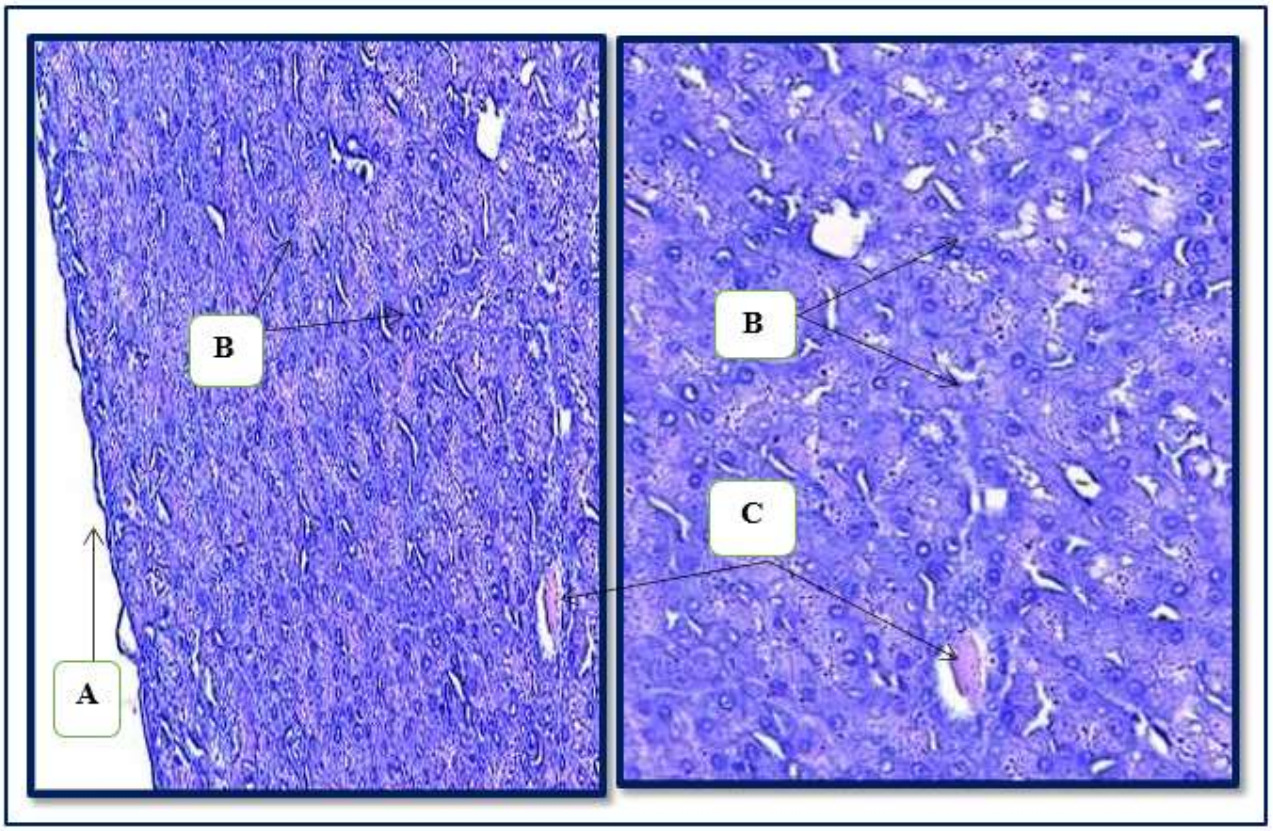

Fig.5. Photographic of the pancreas in francolin show the: **A**. Capsule, **B**. Mucin, **C**. Intercalated duct.

X200 PAS

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