

Histological Study of Exocrine part of Pancreas in Laying hens (*Gallus gallus domesticus*)

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Abstract:

The histological structure of the exocrine pancreas was examined in the current study for fifteen healthy adult female Laying hens. The study showed the pancreas was serous tubuloacinar gland, which has both an exocrine and an endocrine component, the parenchyma of the gland was encircled by a thin layer of loose connective tissue capsule. The exocrine part formed of acini (secretory unites) and excretory ducts. In the current study, the shape of the acini was ovoid, rounded or elongated, were lined by a single layer of pyramidal to columnar cells. The duct system started from centroacinar cells and was organized as intercalated ducts, intra lobular ducts, inter lobular ducts, and main pancreatic ducts. The intercalated ducts were lined by single layer of simple squamous, intra lobular ducts was encircled by simple cuboidal cells, interlobular ducts was lined with simple columnar epithelium and the main ducts were lined with simple columnar epithelial cells. The pancreas of Laying hens was identical to other bird species, but with a few minor species.

Keywords: Pancreas, Histology, Exocrine, Laying hens.

Introduction

Chickens are among the most common and widespread pets, with a total number of hens it 23.7 billion. The commercial Laying hen industry's primary objective is to produce eggs in sufficient numbers to meet public needs, and its composition is primarily determined by hen breed, age, and nutrition [1]. The majority of Laying hens are fed a full diet that has been professionally designed to offer all the nutrients required for optimum health, egg production and welfare, like seeds, soya bean, energy for production daily egg, and proteins [2, 3]. The pancreas of birds is a separate, essential gland with double function and is a type of compound tubuloacinar gland with serous secretion. The exocrine was designed in the shape of a serous tubuloacinar gland, which containing acinus and excretory ducts that are accountable for secretes of digestive juice (hydrolytic enzymes) and electrolytes in to the duodenum [4, 5]. Each acinus was round, oval or elongate in shape and is made up of a small group of secretory low columnar epithelial cells (acinar cells), these cells have a round, large, dark and basally to centrally nucleus site with prominent nucleoli [6]. The cytoplasm of the acinar cells are rich with secretory zymogene granules that fill the apical part of these cells [7]. Pancreatic ducts run from the acini to the point where the organ empties its contents

(acini products) into the duodenum in the following order: intercalated ducts, intra lobular ducts, inter lobular ducts and pancreatic ducts (main ducts) which are two, dorsal and ventral ducts [8, 9]. The cells lining the ducts converted gradually from flattened or low cuboidal in the intercalated duct to cuboidal in the intra lobular duct, while in the interlobular and main ducts, they become simple columnar cells [10]. Three layers make up the main pancreatic duct wall in poultry: mucosa is the first, muscular is the second, and the last adventitia layers [11]. The goal of the current study was to discover the exocrine tissue of the pancreas histologically in Laying hens.

Materials and Methods

For the current investigation, pancreatic samples were taken from 15 healthy adult female Laying hens. Every laying hens was slaughtered. Directly, the abdominal feathers were removed and a longitudinal ventral incision was made in the body cavity, after which the abdominal membranes were removed and the duodenum loop was extracted. Then pancreas was removed, were divided into approximately 4 equal parts. The resulting tissues were washed by 0.9% normal saline (sodium chloride). All part of pancreatic lobes was transferred to fixative included 10% formalin for 24 hours, the samples are rinsed with tap water for (4-5) hours, then dehydrated by passing it through ascending sequence of ethanol concentration gradients and clearing by xylene, embedding in a molten paraffin wax. Tissue blocks were sliced to 4-6 μm , then placed on glass slides. Sections were stained with H and E stain, APS stain and Masson trichrom stain, as well as Gomori's stain. Using a light microscope equipped with digital camera, photographs of the investigated slides were taken. Using an ocular micrometre, the diameter of the acini, the height of the acinar cells, and the thickness of the capsule were all measured.

Results and Discussions

In the present study, the Laying hens pancreas was encircled by a thin layer of loose CT (connective tissue) capsule made primarily of collagen, elastic and reticular fibers as well as fibroblasts. The capsule was lined with flattened mesothelium cells as illustrated in (Figure 1). The outcome of the capsule is consistent with [11] in native ducks (*Anus platyrhynchos*), [12] in Caspian gull, [13] in turkey, [14] in chick pancreas and [15] in kestrel. On the other hand, these result was in contrast with [16] in turkeys and [17] in Ostrich (*Struthio camelus*), they said that a thick, double-layered capsule with an inner fibrous tissue enclosed the pancreas. The histological results demonstrated the average thickness of this capsule in Laying hens were ($11.95 \pm 0.94 \mu\text{m}$) as shown in (Table 1). As a result of this study, the average body weight of adult female Laying hens is 2200gm and average weight of whole pancreas is 4.22gm, and the percent of the pancreas weight to body weight is about 0.20 %.

The study revealed that the parenchyma of the pancreas were mainly composed of an exocrine and endocrine portions. The tissue of pancreas indicated that the exocrine portion occupied larger area in comparison with that of the islets of Langerhans, as showed in (Figure 2). That is comparable to the results with [18] in the red jungle fowl, [19] in the pigeon (*Aquila Chrysaetos*) pancreas. The result appeared the exocrine part formed of acini (secretory unites) and excretory ducts with different diameters as shown in (Figure 3). This finding in agreement with previous studies like [20] in white eared bulbul (*Pycnonotus leucotis*), [12] in Caspian gull, [21] in falcon (*Falco peregrinus*) and [22] in pigeon (*Columba livia*). The shape of the acini in the current work which were ovoid, rounded or elongated in shape and lined by a single layer of pyramidal to columnar cells that sit on the basement membranes, which contain a spherical nucleus in the basal region of the cells. One or more nucleoli may also be visible, zymogen granules were located at the apical portion of these cells as showed in (Figure 4). This results concords with [9] in common gull, [23] in Japanese Quail and [24] in palam Dove. The acinar cells had bizonal cytoplasm, which indicated to the appearance of acidophilic granules in the apical area and rich in mitochondria and

endoplasmic reticulum (ER) in the basal area, which in line with [25] in early hatched goose and [26] in golden eagle, whom found that the reason of the pancreatic acini cells appear bizonal character to the existence of zymogenic granules and mitochondria in the apical and basal regions, as well as the presence of cytoplasm with eosinophilic zymogen granules. The results revealed that centroacinar cells were present in the lumen of the acini unit. In comparison to acinar cells, these cells had a flattened or rounded form, were smaller and brighter and lacked zymogen granules. They also had noticeable round dark nuclei with pale cytoplasm as (Figure 4, 5). The findings of the current investigation on centroacinar cells is in agreement with [24] in the pancreas of palam Dove, [27] in the pigeon, [15] in Kestrel and [28] in chabro chicken. But, the centroacinar cells were not found in Caspian gull by [12], Mynah by [29] and in Common gull and Guinea fowl by [9]. In the present study, the average acini cells height was ($12.03 \pm 0.96 \mu\text{m}$), while the average acini unit diameter was ($29.75 \pm 1.26 \mu\text{m}$) as noted in (Table 1).

According to the results of this study, the duct system started from centroacinar cells and was organized as intercalated ducts, intra lobular ducts, inter lobular ducts, and main pancreatic ducts. The intercalated ducts in hens formed the extension to centroacinar cells by having a part of intra-acinar lined with centroacinar cells, which is represented as a single layer of simple squamous epithelium as showed in (Figure 6). These findings has been reported in the pancreas of Kestrel (*Falco tinnuclulus*) by [15], in pancreas of falcon by [30] and in chabro chicken by [28], they stated the exocrine pancreatic ducts began with intercalated duct which were lined with squamous epithelium. These results disagreement with the results of [19] in pigeon whom stated that the intercalated duct was enveloped by simple cuboidal cells. Also, [7] in adult Kestrel found that the intercalated duct was not found. The intra lobular ducts was encircled by simple cuboidal cells with rounded nuclei, were surrounded by CT (connective tissue) fibers with few elastic fibers are found surrounding these channels. The ducts lumen was filled with secretory material as illustrated in (Figure 7). The results of the present observation are in line with [31] in duck, [32] in goose and [33] in Quail and Cattle Egret. The result disagreement with [8] in goose, who discovered the intra lobular ducts were bordered by columnar epithelium and with [30] in falcon, who identified that simple squamous epithelium enveloped the intra lobular ducts. The histological results indicated the interlobular ducts in Laying hens was lined with simple columnar epithelium and bounded with CT (connective tissue) fibers (collagen fibers with few elastic fibers and smooth muscle fibers). The ducts lumen was filled with secretory material as showed in (Figure 8). These findings are in agreement with [18] in red jungle fowl, [26] in golden eagle and [34] in *Coturnix coturnix* and *Pterocles alchata*, whom stated that the interlobular ducts were lined with simple columnar epithelium. While in Kestrel (*Falco tinnuclulus*) by [15] the outcomes showed that the inter lobular duct was found in the connective tissue between the pancreatic lobules, was bordered by a simple columnar to low columnar epithelium, and was surrounded by an abundance of connective tissue. Moreover, [35] in quail (*Coturnix coturnix japonica*) whom stated that the interlobular ducts were lined with a low columnar epithelium, put disagree with [20] in white eared bulbul who revealed that the interlobular ducts enclosed by with simple cuboidal epithelium. The results in the present study noticed there were no serous glands in the wall of the interlobular ducts. This findings concords with results of [12] in Caspian gull, [22] in pigeon and [36] in Mynah. As well as not in line with [33] in Cattle Egret and [10] in goose, whom reported the presence of serous glands in the wall of the interlobular ducts. This findings concords with results of [12] in Caspian gull, [22] in pigeon and [36] in Mynah. As well as not in line with [33] in Cattle Egret and [10] in goose, whom reported the presence of serous glands in the connective tissue of the wall of interlobular duct. The histological findings showed that the interlobular ducts extended into the larger ducts carrying eosinophilic secretory products, which were main ducts. The main ducts then drained into the duodenum. In the current study, simple columnar epithelial cells with rounded nuclei lined the main excretory ducts, and the mucous membrane had arrangement of longitudinal folds. In addition to this, connective tissue fibers were

surrounding these ducts had smooth muscle fibers was thicker in the main ducts as showed in (Figure 9). These results are correlate with [18] in red jungle fowl, [37] in goose, [33] in quail (*Coturnix coturnix japonica*) and Cattle Egret, and [38] in ducks (*Anus platyrhncchos*) and turkey (*Meleagris gallopavo*), and [39] in turkey (*Meleagris gallopava*). Moreover, [29] in Mynah and [32] in goose, the main duct was covered with stratified cuboidal to stratified columnar cells.

Table (1): Measurements of the female adult Laying hens capsule thickness, acini diameter, and acini cell height (Mean $\pm$ Standard deviation).

Pancreatic Measurements / μm (Mean $\pm$ SD)	Laying Hens
Thickness of capsule	11.95 $\pm$ 0.94
Diameter of acini	29.75 $\pm$ 1.26
Height of the acini cells	12.03 $\pm$ 0.96

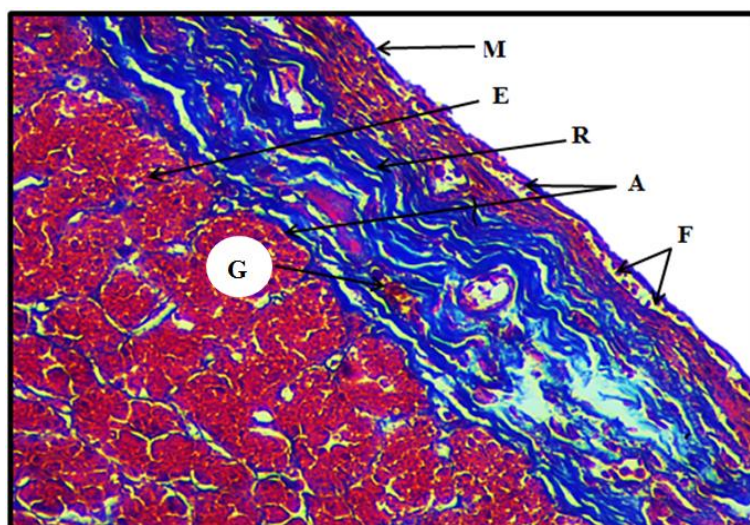


Figure (1): Photomicrograph show the capsule of female adult Laying hens. A- Capsule. R-Connective tissue fibers. F- Fibroblast. E- Parenchyma .M- mesothelium layer. G- Blood vessels. Masson trichrom. stain (200 X).

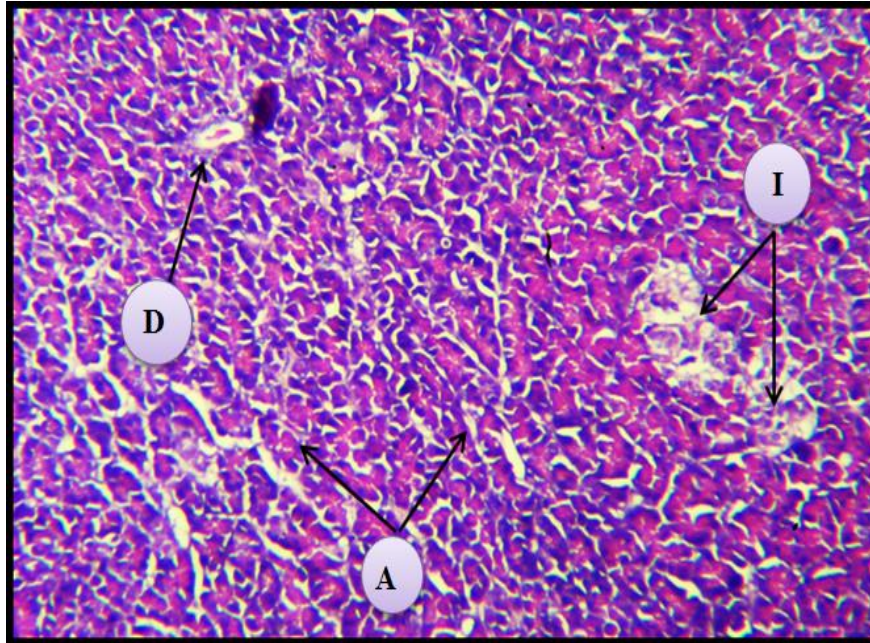


Figure (2): Histological picture of the Laying hens pancreas shows the pancreatic parenchyma, which includes: 1- **Exocrine part** (A- Acini tissue. D- duct system) 2- **Endocrine part** (I- Islets of Langerhans). H&E stain. (100X).

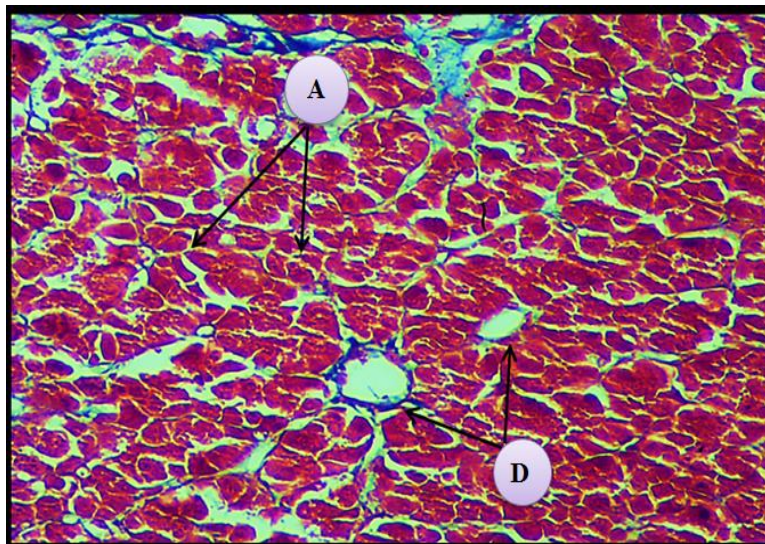


Figure (3): Histological picture of a cross-section in the Laying hens pancreas shows the pancreatic Exocrine part, which includes: A- Acini tissue. D- duct system with different diameters. Masson stain. (200X).

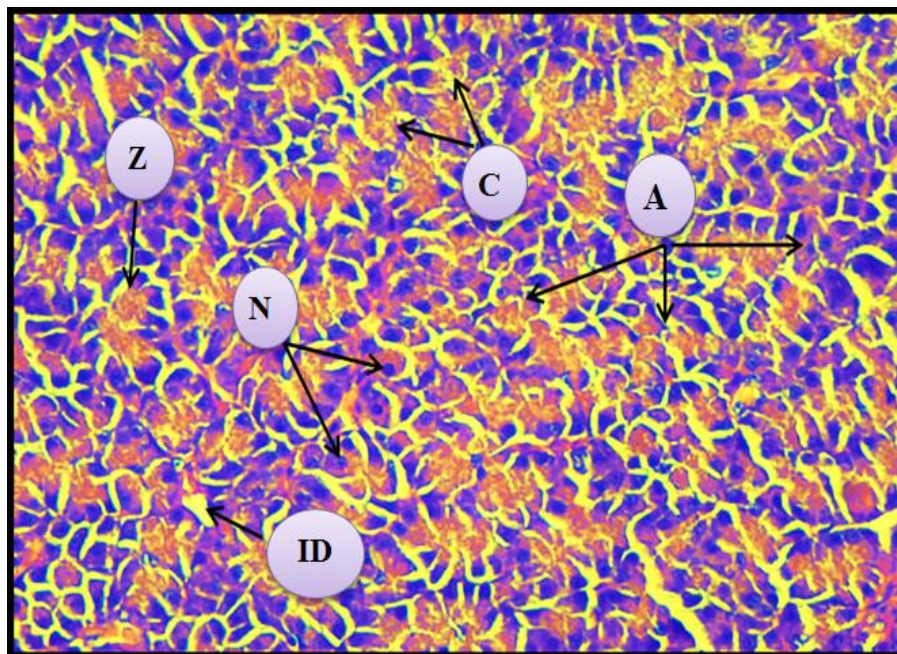


Figure (4): Photomicrograph of a pancreatic Laying hens show the acini tissue, **A**- Acinus. **N**- Nucleus of acinar cells. **Z**- Zymogen granules. **C**- Centroacinar cells. **ID**- Intercalated duct. H&E stain. (200X).

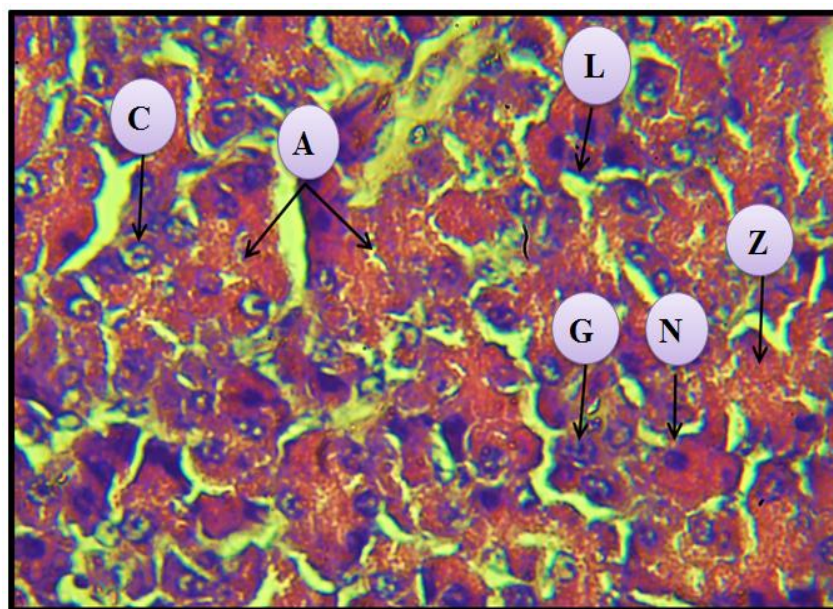


Figure (5): Photomicrograph of a pancreatic Laying hens pancreas show the acini tissue, **A**- Acinus. **N**- Nucleus of acinar cells. **Z**- Zymogen granules. **C**- Centroacinar cells. **G**- Nucleolus of acinar cells. **L**- Acinus lumen. H&E stain. (400X).

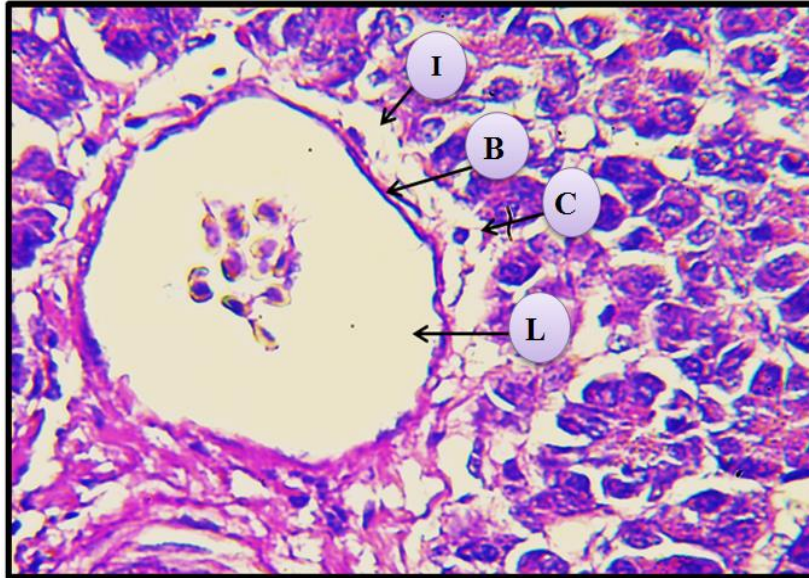


Figure (6): Photomicrograph of a cross-section in the Laying hens pancreas showed, **I**- Intercalated duct. **B**- flat epithelial cells (squamous epithelium) lining the duct. **L**- Lumen of intercalated duct. **C**- centroacinar cells in the pancreatic duct. PAS stain (400X)

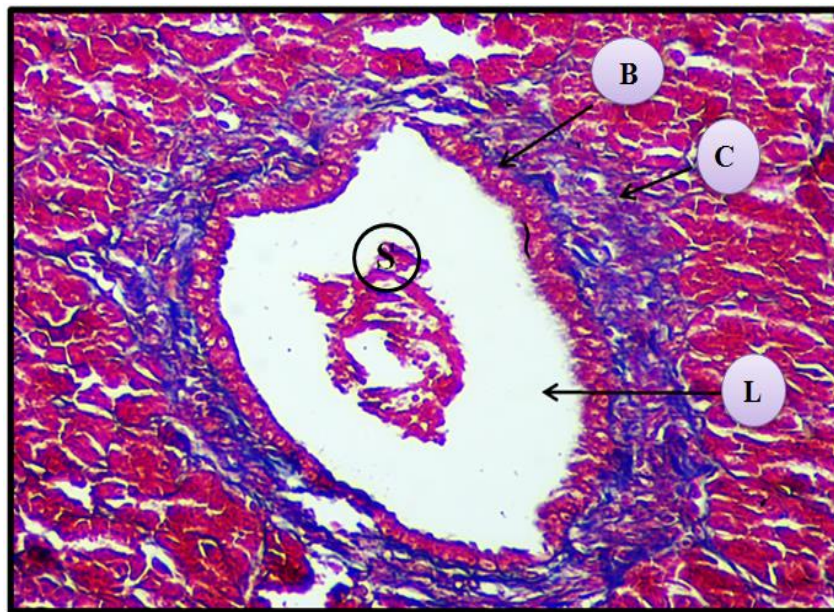


Figure (7): Photomicrograph of a cross-section in the Laying hens pancreas showed, **I**- Intra lobular duct. **B**- epithelial cells (simple cuboidal). **C**- connective tissue fibers. **L**- Lumen of duct. **S**- Acini secretion. Masson stain (200X)

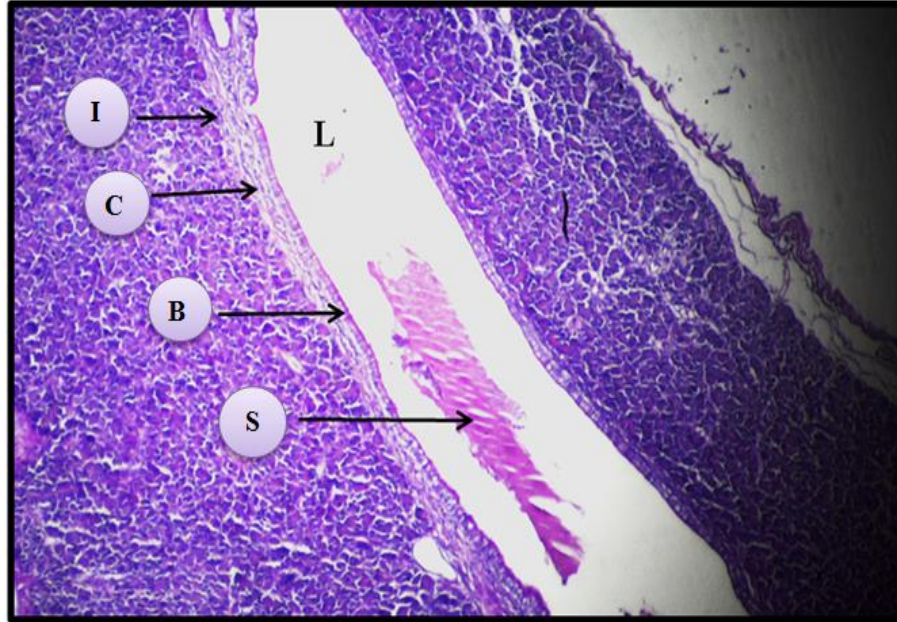


Figure (8): Photomicrograph of a pancreatic Laying hens pancreas showed, **I-** Interlobular duct. **B-** columnar epithelial cells . **C-** connective tissue fibers. **L-** Lumen of duct. **S-** Acini secretion. PAS stain (40X)

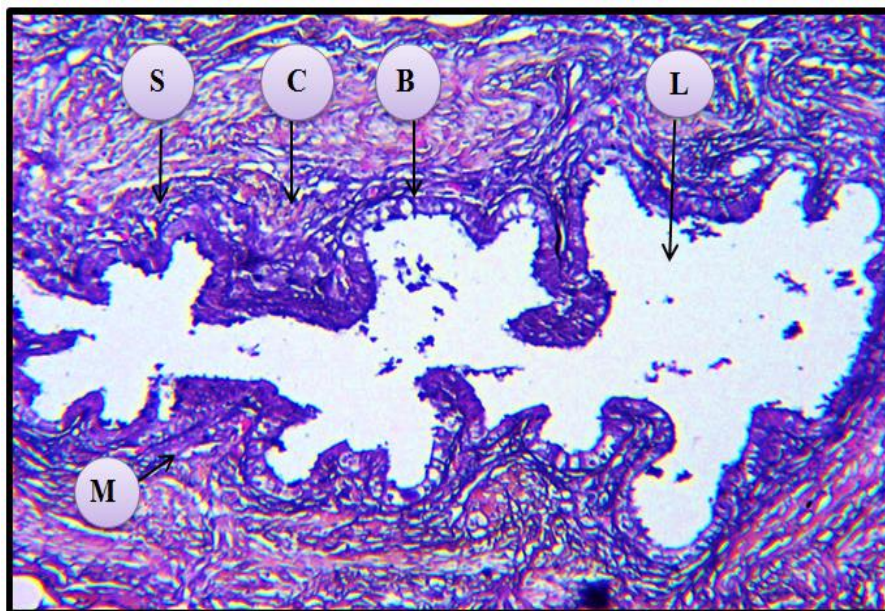


Figure (9): Photomicrograph of a pancreatic Laying hens pancreas showed, **M-** Main duct. **B-** simple columnar epithelium . **C-** connective tissue fibers. **L-** Lumen of duct. **S-** smooth muscle fibers. Gomori's stain (100X)

Conclusions

The pancreas of Laying hens was situated between the descending and ascending loops of the duodenum on the right side of the abdominal cavity, and encircled by a thin layer of loose CT (connective tissue) capsule made primarily of collagen, elastic and reticular fibers as well as fibroblasts. The capsule was lined with flattened mesothelium cells. The parenchyma of the pancreas was mainly composed of exocrine and endocrine portions diffuse between the acini, which symbolize to the islets of Langerhans. The exocrine part of the pancreas was consisted of a serous compound tubuloacinar gland formed of acini with ovoid, rounded or elongated in shape. The acini in this bird were lined by a single layer of pyramidal to columnar cells which contain a spherical nucleus in the basal region of the cells. The acinar cells were had bizonal cytoplasm, which indicated to the appearance of acidophilic granules (Zymogen granules) in the apical area and rich in mitochondria and (ER) endoplasmic reticulum in the basal area. The large diameters of acini and height of acinar cells in hens, due to the high activity of the acini to secrete pancreatic enzymes. In poultry species the secretion of some pancreatic enzymes which are related to bird food components. The ductal system of the exocrine part of Laying hens pancreas was started from centroacinar cells and was organized as intercalated ducts, intra lobular ducts, inter lobular ducts, and main pancreatic ducts.

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