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Histological and Histochemical description of the pancreas in Gallus gallus Domesticus

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***Abstract*—** In birds, the pancreas is crucial for both endocrine and digestive control. Understanding the avian pancreas' glandular makeup and functional secretion patterns is aided by histological characterization. The purpose of this study was to examine the histological architecture of the pancreas in Gallus gallus, with a focus on the identification of exocrine and endocrine components, glandular organization, and secretion types. Standard histology procedures were used for the collection and processing of pancreatic samples from five Gallus gallus domesticus. To examine cellular and glandular features under a microscope, tissues were fixed, embedded in paraffin, sectioned, and stained using histological dyes. A well-developed exocrine section made up of acinar cells with granular cytoplasm suggestive of serous discharge was seen upon histological inspection. the islets with different cellular distributions served as a representation of the endocrine component. Mixed glandular activity and distinct endocrine and exocrine area differentiation were noted in the study. The studied Gallus gallus species' pancreas exhibits a complicated histological architecture with two functioning components. These results serve as a guide for next comparative research and advance anatomical and physiological knowledge of hormone and digestion regulation.

***Keywords* —** Gallus gallus, pancreas, Histological study.

INTRODUCTION

Originating from the Southeast Asian junglefowl during the First Agricultural Revolution, the domestic chicken (Gallus gallus domesticus) is now a common bird all over the world. Charles Darwin believed that only red junglefowls (Gallus gallus) were used to domesticate hens. It was suggested that four species of junglefowl—the red junglefowl, which is indigenous to Indochina, the gray junglefowl (Gallus sonneratii), which is native to the Indian subcontinent, the Ceylon or Sri Lanka junglefowl (Gallus lafayetti), and the green junglefowl (Gallus varius)—were the ancestors of modern chickens (1). The alimentary tract is related to the pancreas, a big gland. The pancreas is situated between the duodenum's two limbs on the right side of the abdominal cavity in all birds. As in chickens and quail, the pancreas of birds is thought to contain four lobes: ventral, dorsal, third, and splenic; additionally, it has three ducts: ventral, dorsal, and third (2). The most significant differences between the various species of the avian pancreas are the division of lobes, distribution of the islets within lobes, the form and frequency of endocrine cells in islets, and the structure of ducts. The avian pancreas is different from other animals in that it is composed of two or more lobes and contains two types of islets (3).

Birds' pancreas is a form of complex tubuloacinar gland that secretes serous fluid and is a separate, vital gland with two functions. The exocrine was fashioned after a serous tubuloacinar gland, which has acinus and excretory ducts that are responsible for secreting electrolytes and digesting juice (hydrolytic enzymes) into the duodenum (4, 5). Composed of a small group of secretory epithelial cells (acinar cells), each acinus was round, oval, or elongated in shape. These cells feature a large, round, dark nucleus site that is located from the base to the center, with conspicuous nucleoli. (6).

The apical region of the acinar cells is filled with secretory zymogene granules that are abundant in their cytoplasm (7). Intercalated ducts, intralobular ducts, interlobular ducts, and pancreatic ducts (main ducts), which are two dorsal and ventral ducts, lead from the acini to the place where the organ empties its contents (acini products) into the duodenum (8, 9). While the cells lining the ducts became simple columnar cells in the interlobular and main ducts, they eventually changed from flattened or low cuboidal in the intercalated duct to cuboidal in the intralobular duct (10). The primary pancreatic duct wall in chicken is composed of three layers: the first is the mucosa, followed by the second is the muscle, and the final layer is adventitia (11).

The exocrine and endocrine functions of the pancreas are carried out by distinct cell groups, and the organ's exocrine and endocrine functions are both supported by certain cells (12). As stated by, the islets were dispersed at random throughout the exocrine portion of each lobe. (13) In guinea fowls and common gulls, however, they were more concentrated in the central region of the parenchyma than in the peripheral regions, especially in the splenic lobes, where they frequently made up the sizable central mass of endocrine tissue.

Light-colored oval and irregular cells clumped together to constitute the majority of the pancreatic islets of Langerhans. We discovered two kinds of islets, namely. They were light islets made up of both mixed islets with both dark and light cells and light cells (14, 15, 16, 17).

In Gallus gallus, histological details of the pancreas are currently unavailable. Consequently, this study sought to illustrate the histological and histochemical composition of its pancreatic gland.

MATERIALS AND MTHODS

For this study, pancreatic samples were obtained from five laying hens. The abdominal membranes were removed along with the duodenal loop. The pancreas was chopped and submerged in a fixative containing 10% formalin for an entire day. Each pancreatic lobe piece was then washed with xylene, gradually dehydrated by increasing the ethanol concentration, and embedded in melted paraffin wax. Tissue samples were sliced to 5 μm and then put on glass slides (15).

Sections were stained using H and E stain, Gomori's stain, Masson trichrome stain, and PAS stain. A light microscope with a digital camera attached was used to take pictures of the analyzed slides. An ocular micrometer was used to measure the thickness of the capsule, the diameter of the acini and diameter of islets of langerhans.

Ethics-approved

This study was conducted in the anatomical laboratory of the University of Kerbala's College of Veterinary Medicine under reference number UOK.VET.AN.2025.127.

RESULT AND DISCUSSION

Histological descriptions of pancreas of Gallus gallus

In Gallus gallus (chicken), the pancreatic histology shows an extraordinarily ordered glandular arrangement backed up by a delicate yet extensive stromal framework that gives the gland both mechanical assistance and a vascular nourishment. The main exocrine component of the gland is made up of the stromal tissue, which is made up of connective elements that divide the parenchyma into separate lobes. Within the various lobes, there are numerous pancreatic acini.

These acini are organized as groups of pyramidal epithelial cells, each of which has an irregular nucleus at the base and cytoplasm that is oriented apically and contains zymogen granules, indicating that it secretes proteins. Because the avian pancreas plays a crucial role in exocrine activity, the acini work together to synthesize and secrete digestive enzymes (Fig,1). Unevenly shaped clusters of endocrine tissue known as the islets of Langerhans are seen scattered throughout the exocrine lobules.

These pancreatic islets seem irregular and dispersed throughout the lobular parenchyma due to their varied contours rather than their uniform shape. Particularly noteworthy is the cellular organization found within the islets, where two main cell populations can be identified based on staining properties and morphology. While irregularly shaped, dark-staining cells are concentrated toward the center of the islets, rounded, pale-staining cells are primarily found at their periphery.

The present arrangement provides insight into the biological heterogeneity of the islets. It is compatible with the recognized endocrine cellular composition of the avian pancreas, where pale-staining cells are linked to the generation of insulin, while darker-staining cells were associated with populations that secrete glucagon and somatostatin. The exocrine and endocrine compartments' intimate relationship digestive functions are combined into a single organ system (Fig, 1-3).

The ductal system

The remarkable development of the avian pancreas reflects the need for efficient secretory substance transport into the intestinal tract. At the smallest scale, the intercalated ducts are visible. They are encircled by a simple squamous epithelium that forms a delicate interface with the acinar units.

These ducts serve as the first connection of the ductal system and gather discharge straight from the acini. As the duct capacity increases, the simple cuboidal epithelium that lines the interlobular ducts—which progress toward larger channels—provides a more robust lining than the squamous lining of intercalated ducts. The ducts are located within the connective tissue septa that divide lobules.

Along with the ductal and parenchymal components, the pancreatic vascular supply is significant and essential to its operation in Gallus gallus. Distributed throughout the stromal tissue, the pancreatic arteries offer a vast vascular network that guarantees sufficient oxygen and nutrient delivery and speeds up the passage of endocrine secretions into the systemic circulation.

Overall, the current evaluation of the pancreas in Gallus gallus reveal a well-balanced organ that integrates (exocrine and endocrine) activities within a well-structured stromal and bloodstream structure. A continuously distinct draining system removed the acini, which were grouped into clusters and supplied the digestive enzymes required for effective mineral decomposition. system. Conversely, the asymmetrical islets of Langerhans regulate the process of metabolism and systemically blood glucose levels. They consist of masses of cells with different morphologies that are distributed in a gradient from the periphery to the center (Fig. 3-6).

Histochemical study

The arrangement of mucous substances and carbohydrate in Gallus' pancreas cells was assessed using (PAS) staining. Acinar particles, drainage structures, and basement wall membranes were all prominently displayed by the response, therefore enabling structural evaluation. The present study finding the pancreas in these birds exhibited a robust positive (PAS) response in Gallus. Granules staining prominently and the acinar cells' intense magenta coloring suggested a high concentration of muco-substances and glycogen. The abundance of (PAS) reactive material was notable positivity shown by ductal cells and the basement membranes that are connected to them; strong staining pattern indicates that the exocrine pancreas has an active secretory profile and that carbohydrates are stored robustly. (Figure 7).

The spatial arrangement and percentage of collagen fibers in the pancreatic stroma were assessed using Masson's trichrome stain. The structural investigation was made possible by the stain's ability to clearly distinguish connective tissue

components from the surrounding tissue matrix. Collagen fibers were prominently deposited , especially around the network of ducts and blood supply The corresponding fibers, which formed a dense supporting support structure around blood vessel and drainage structures, were conspicuously displayed in blue. Conversely, there was very little collagen in the stromal tissue connected to the acini, demonstrating that the accumulation of collagen was mostly limited to the surrounding vascular and periductal areas. The overall structure points to a strong structure of connective tissue that provides stability to the draining elements of the pancreas. (Figure8). Gomori Staining: The fundamental arrangement of the pancreatic Gallus was assessed using Gomori's stain, with special attention paid to the c framework, acinar and Langerhans structure . This approach allowed for visual inspection of the sustaining stromal aspects and the distinct separation of the pancreatic components. langerhans that were clearly separated from adjacent extracellular tissue.

Comprehensive cellular differentiation and arrangement into tight rectangular forms were evident in the easily visible acinar cells. Additionally, visible became the construction of connective tissue, which separated acinar components into discrete lobules and provided building support. Collagenous elements within the stroma were highlighted, outlining both vascular and ductal structures. The clarity of staining in Gallus reflects a well-organized pancreatic architecture with a strong distinction between endocrine and exocrine compartments (Fig 10).

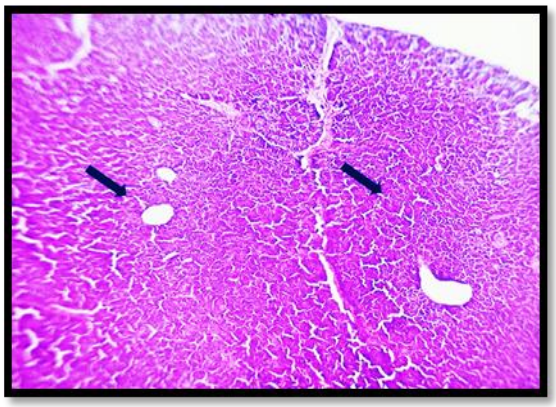

Figure 1. Photography of Pancreas (G) show the pancreatic stromal tissue , pancreatic lobes distributed the pancreatic acini (black arrow).H&E stain.100X.

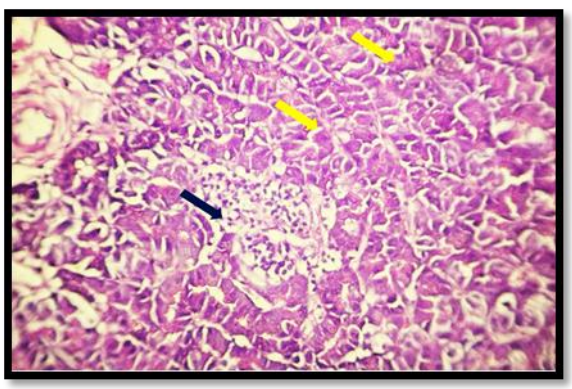

Figure 2. Photography of Pancreas (G) show the islet of Langerhans irregular in shape (black arrows) and pancreas acini (yellow arrows). H&E stain, 400X.

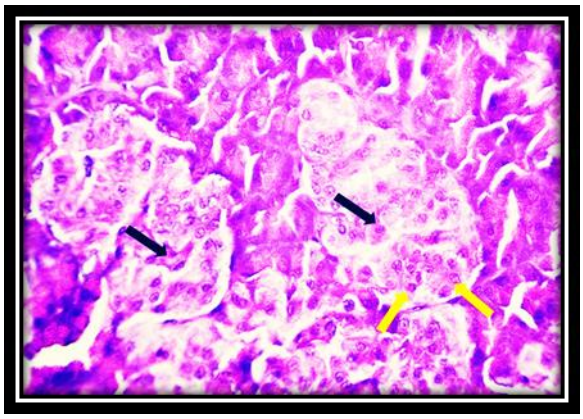

Figure 3. Photography of Pancreas (G) show the islet of Langerhans had two types of cells rounded-pale cells located peripherally (yellow arrows) and irregular-dark cells located in center (black arrows). H&E stain. 400X.

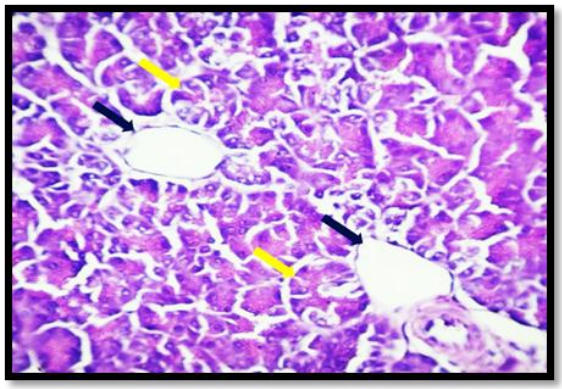

Figure 4. Photography of Pancreas (G) show the intercalated duct lined by simple squamous epithelia (black arrows) and exocrine acinar cells had irregular nucleus located in base of cells (yellow arrows). H&E stain. 400X.

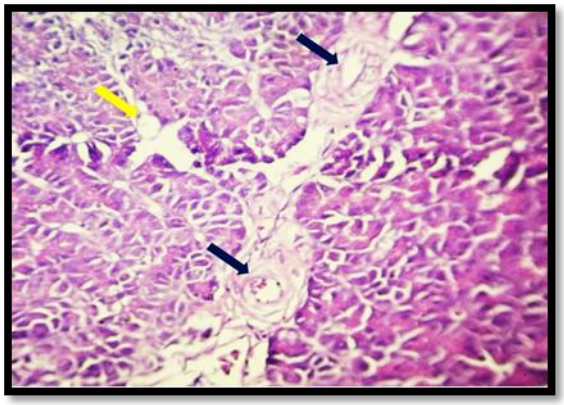

Figure 5. Photography of Pancreas (G) show the well development ductal system, interlobular duct lined by simple cuboidal epithelia (black arrow), the intercalated duct (yellow arrow). H&E stain. 400X.

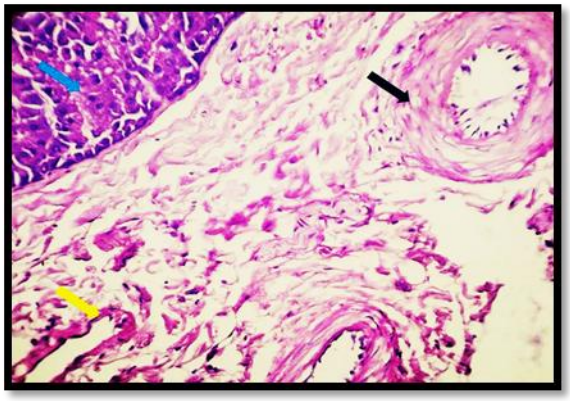

Figure 6. Photography of Pancreas (G) (blue arrow) show the main duct lined by stratified cuboidal epithelia (yellow arrow). Pancreatic artery which supplied pancreatic stroma (black arrow). H&E stain. 400X.

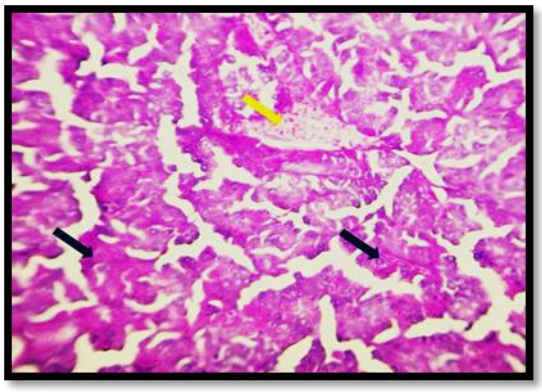

Figure 7. Photography of Pancreas (G) show the strong positives reaction highlights glycogen and mucosubstances, revealing magenta acinar granules, ducts, and basement membranes for structural evaluation. PAS stain. 400X.

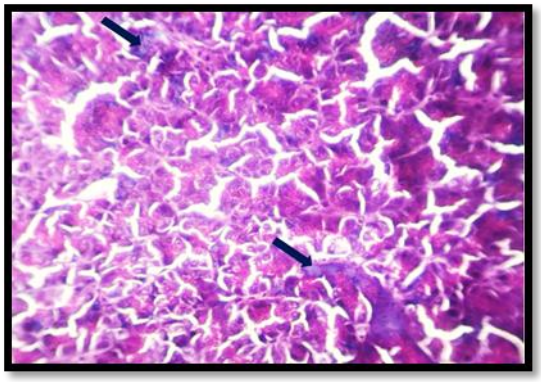

Figure 8. Photography of Pancreas (G) show the pancreatic stromal tissue had scant amount of collagen fibers surrounded the pancreatic acini (black arrow). Masson trichrome stain.400X.

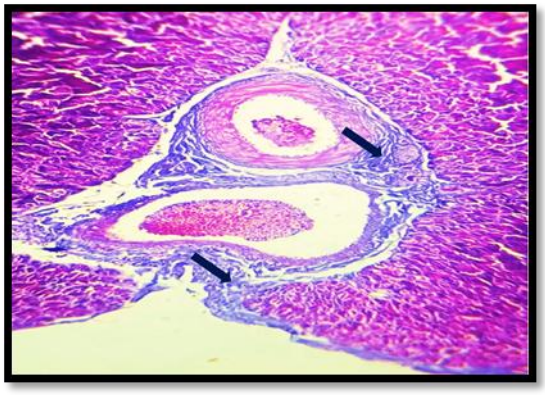

Figure 9. Photography of Pancreas (G) show the a large amount of collagen fibers surrounded the blood supply vessels and ductal system (black arrows). Masson trichrome stain 400X.

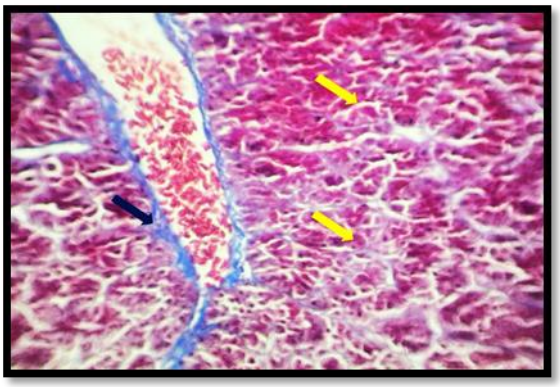

Figure 10. Photography of Pancreas (G) show histological pancreatic tissue with Gomori stain well-defined islets, distinct acinar cells (yellow arrow), and clear connective tissue framework (black arrow). Gomori stain. 400X.

The fragile sheath of loose connective tissue capsule that encased the pancreas of the gallus in this study was primarily made up of collagen, elastic, and reticular fibers, as well as fibroblasts. Mesothelial cells were flattened and applied to the capsule. The capsule's findings are consistent with (11) in wild ducks (Anas platyrhynchos),(18) in Caspian gulls, (19) in turkeys, (20) in chick pancreases, and (21) in kestrels. Conversely, these findings differed from those in (22) for turkeys and (23) in Ostrich (Struthio camelus), It was stated that the pancreas is surrounded by a dense, double-layered capsule containing inner fibrous tissue. The research showed that the pancreas's parenchyma primarily consisted of exocrine and endocrine sections. The pancreatic tissue showed that the exocrine part covered a greater area than the islets of Langerhans. That is similar to the outcomes with (24) in the red jungle fowal, (25) in the pigeon (Aquila Chrysaetos) pancreas. The outcome showed that the exocrine section was made up of acini (secretory units) and excretory ducts of varying diameters. This result aligns with earlier research such as (26) in white-eared bulbul (Pycnonotus leucotis), (12) in Caspian gull, (27)in falcon (Falco peregrinus) (28), and in pigeon (Columba livia). In the current investigation, the acini were ovoid, rounded, or elongated, with a single layer of pyramidal to columnar cells on

the basement membranes and a spherical nucleus in the cell's base. Zymogen granules are located at the apex of these cells, and one or more nucleoli may also be seen (33). For the common gull (9), for the Japanese quail (29,30) for the palam dove are all consistent with this result.

Bizonal cytoplasm was seen in the acinar cells, indicating the presence of many mitochondria and acidophilic granules in the apical region. The results of this study show that the duct system was formed by centroacinar cells and had the following structures: intercalated ducts, intralobular ducts, interlobular ducts, and the main pancreatic ducts. An intra-acinar segment lined with centroacinar cells, represented as a single layer of simple squamous epithelium, was included in the intercalated ducts in hens to extend to centroacinar cells. These outcomes have been reported in the pancreas of falcons by (32), kestrels (Falco tinnunculus) by (21) and chabro chicken by (31). They stated that intercalated ducts made of squamous epithelium served as the precursors of the exocrine pancreatic ducts. These findings go counter to those of (25) in pigeons, which showed that simple cuboidal cells encircled the intercalated duct.

Furthermore, the intercalated duct was not seen in adult Kestrels (7). Simple cuboidal cells with rounded nuclei surrounding the intralobular ducts. Connective tissue (CT) fibers bordered the channels, and a few elastic fibers were present as well.

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