

Histochemical Studies of Pancreatic Disorders in Chabro and Vanraja Chicken

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ABSTRACT

The pancreas is a highly specialized multitasking organ, which plays a pivotal role in poultry production. The extent of pancreatic pathology in different poultry diseases of economic importance at different age groups and breeds particularly in Chabro and Vanaraja breeds of poultry has been studied to evaluate the lesions of pancreas in different pathological conditions and its correlation with various poultry diseases. An aggregate of 240 pancreases were gathered from chicken affected from different viral, bacterial, protozoal, fungal, metabolic, and toxicological diseases. The disease was confirmed by necropsy findings and further analysed by histochemistry with special staining. The major histopathological changes in pancreas in decreasing order of frequency were interstitial fibrosis, congestion, individualization of acinar cells, pancreatic fat necrosis in exocrine pancreas, periductular fibrosis and capsular thickening. However, no direct correlation was found between gross pathology and histopathological findings. It has been observed that pancreatic disorders were more common in metabolic and chronic diseases as compared to other infectious poultry diseases. Significant pancreatic pathology and their dysfunctions are an important contributory factor in chronic disease of known or idiopathic origin. Supplementation of pancreatic enzymes in poultry ration in case of disease outbreak/poor growth performance are suggestive for optimal functioning of pancreas and in turn assurance of healthy poultry flock.

Keywords: Pancreas, Histopathology, Histochemistry, Vanaraja and Chabro.

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INTRODUCTION

The pancreas in poultry is collection of three elongated lobes viz dorsal, ventral and splenic, each of which empties the content of digestive enzymes through separate duct in the terminal part of ascending duodenal loop alongside bile duct and hepatic duct. It is relatively small in carnivores and granivores but larger in picivores and insectivores. In duck it is quite small extending upto 3/4th of duodenal loop. In mammals on the other hand, pancreas is flattened, lobulated and pink or grey tubuloalveolar gland, a large portion of which is located in the mesentery immediately adjacent to duodenum. However, the functional units and cellular arrangements of pancreatic parenchyma is quite similar in birds and mammals.

Since the viscera of poultry is not compartmentalized it is easy for the infection to spread in the entire organ system of birds in conditions like mushy chick disease, chick edema disease, egg peritonitis, visceral gout, air sacculitis etc. to name a few. Curving of duodenal loop (J-like appearance) and pancreatic atrophy, has been reported during Postmortem examinations of broiler chicken infected with parvovirus (Nunez *et al.*, 2016) and zinc toxicosis in ostrich as well. It was also found that

major histological lesions were acute pancreatic necrosis and their atrophy due to different strain of influenza viruses in duck (Brojer *et al.*, 2009), swan (*Cygnus cygnus*) (Teifke *et al.*, 2007) and chicks (Shinya *et al.*, 1995). Avirulent Newcastle Disease Virus was also shown to replicate in the pancreas of chickens causing acute pancreatitis. Nakamura *et al.*, (2002) reported pancreatic multifocal necrosis in pancreatic acinar cells in adenovirus-associated hydro pericardium syndrome (Leechi Heart Diseases) of broiler chicken. Pancreatic Adenocarcinoma has been evidenced in guinea fowl and chicken by Okoye *et al.*, (1993) and Abdul-Aziz (1995) respectively. Thus, it has been envisaged to investigate the extent of histopathological alteration in correlation with poultry diseases of economic significance in Chabro and Vanaraja breeds of Chickens.

MATERIALS AND METHODS

The present study, records pertaining to pancreatic disorders of total no.240 dead birds (Chabro and Vanaraja) during the post-mortem examinations conducted at the Department of Veterinary Pathology, Bihar Veterinary College Patna, Bihar

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Animal Science University, Patna, were examined and then tabulated. The information with regard to age, breed weight, types of feed, vaccination, clinical signs etc. were collected as per requisition form submitted to the Department. All precautionary measures were taken to protect our self and environment from infectious diseases, if any. The post-mortem examinations were conducted as early as possible to minimize the putrefaction or autolysis. Cause of death was determined on the basis of necropsy findings. The pancreas of each bird was fixed in 10% neutral buffered formalin for histochemical studies. Fixed tissues were processed by routine paraffin embedding technique (Luna, 1968). The paraffin sections of 5 μ were obtained for different histochemical studies. The sections were stained by Periodic Acid Schiff (P.A.S.) method for demonstration of Zymogen Granules, and Mallory's trichrome stain to demonstrate the presence of various amount of collagen fibre in interstitial region of pancreas.

RESULTS AND DISCUSSION

The major histopathological changes in pancreas in decreasing order of frequency were interstitial fibrosis (54.41% Vanaraja and 46.00% Chabro), congestion (46.00% Vanaraja and 30.00 % Chabro), periductular fibrosis (27.94% Vanaraja and 13.20% Chabro) and capsular thickening (13.24% Vanaraja and 5.39% Chabro). However, the principal histopathological finding was interstitial fibrosis in the cases of pancreatic deformities in general and marked interlobular and periductal fibrosis in particular (Fig. 1). Interstitial fibrosis may play important role in development of pancreatic deformity since collagenous tissue being firm and strong may resist expansive growth of pancreatic parenchyma particularly in growing birds. Mallory's trichrome stain (Fig. 2) was especially used to demonstrate the presence of various amount of collagen fibre in interstitial and periductular region. Mahmood *et al.* (2020) also found it as a perfect staining reaction to visualize collagen fibers in pathological samples in sheep diagnosed with chronic fasciolosis which is similar to that obtained by traditional Masson's trichrome stain. In case of pancreatic injury, the stellate cells undergo myofibroblastic transformation with capability to synthesize fibrillar collagen. (Shek *et al.*, 2002). Which forms the ground work for overt interstitial fibrosis. However, this mechanism in avian pancreatic fibrosis needs further exploration and confirmation.

Congestion was another consistent microscopic change in most of the disease conditions in vanraja and Chabro poultry birds, though there was variation in the degree of hyperaemia. Persistent congestion can bring about hypoxic damage to the parenchymal cells being drained by the veins of the affected pancreatic parenchyma. Hypoxia often leads to cellular swelling, vacuolar degeneration and necrosis. Hypoxia often leads to cellular swelling, vacuolar degeneration and necrosis. These changes were in corroboration with the findings of Jain (2009) and Yadav *et al.* (2020).

The multifocal exocrine pancreatic fat necrosis (Fig. 3) has been registered in all age group of Vanraja and Chabro birds, however their incidence was significantly higher in viral diseases such as Infectious bronchitis (IB), fowl pox, leucosis, chicken infectious anaemia, ranikhet disease and infectious

bursal diseases (IBD) which suggests pancreas is habitually invaded by pathogenic viruses as also reported by Charles (2007). Diffuse pancreatic necrosis is definitely going to reduce the functional status of pancreas, resulting into production loss of the flock.

Thickened pancreatic capsule (Fig. 4) has been observed as an associated lesions with active inflammatory conditions in pancreas which predisposes the organ to adhesive changes with the organ of the digestive system. This had been distinctly observed in cases of lymphoid leukosis or cystadenocarcinoma, where the pancreas becomes completely embedded in thickened capsule. Encapsulation brings about degenerative and necrotic changes in pancreatic acini, especially those which were completely surrounded by fibrous connective tissue. These changes normally are suggestive of obstruction in the network of pancreatic ducture, resulting in reduced excretion of pancreatic enzyme despite being continually produced by acinar cells. These end up in increased zymogen granules concentration in acinar cells (Fig. 6) (Pound *et al.*, 1981).

The cytoplasm of acinar cells and cytoplasmic granules exhibited mild to moderate and intense reaction for Periodic Acid Schiff (PAS), respectively in pancreatic parenchyma of Vanraja and Chabro poultry birds (Fig. 7). Jain (2009) also reported that PAS activity was moderate in chicks of CARI Shyama, mild to moderate in chicks of Vanaraja, moderate in growers of CARI Shyama, intense in growers of Vanaraja and mild to moderate in pullets of both breeds which was in corroboration with our findings. Hamodi *et al.* (2013) and Yadav *et al.* (2020) stated that the pancreatic acinar cells were moderately positive for PAS in Common gull and Guinea fowl. Yadav *et al.* (2020) also stated that the pancreatic acinar cells were moderately positive for PAS in chabero chicken. However, according to Vaish (2005) in Kadaknath fowl negative reaction for PAS was observed in all the structures of pancreas. Increased concentration of zymogen granules in the acinar cell was observed in small percentage of diseased Vanraja and Chabro birds, however this change was quite high in birds suffering from brooder pneumonia. Any deficiency or toxicity, especially zinc toxicity damages the basement

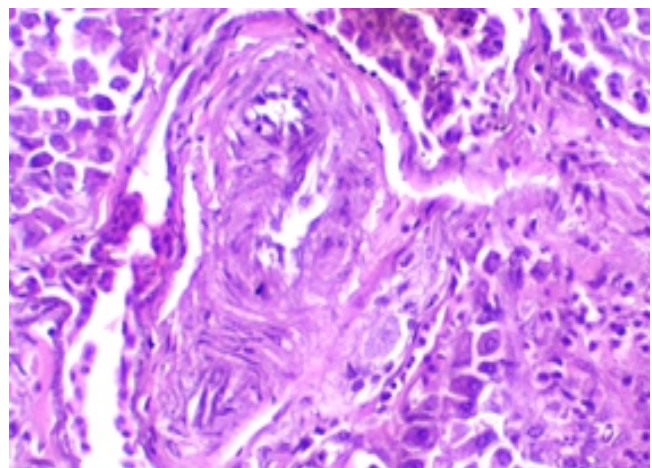


Fig.1: Photomicrograph showing atrophy of exocrine pancreatic glands due to Inter-lobular fibrosis in Chabro Poultry Birds (H&E Stain; 400X).

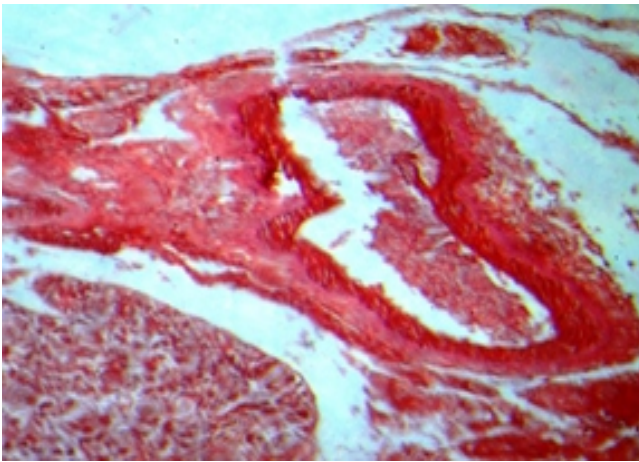


Fig.2: Photomicrograph showing large duct (Yellow arrow) with pancreatic secretions (Red arrow) and marked periductular fibrosis (Green arrow) - Mallory trichome stain 100X

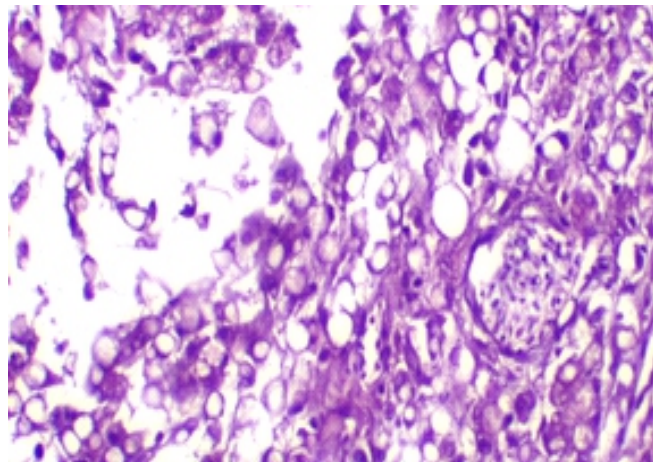


Fig.3: Photomicrograph showing diffuse pancreatic fat necrosis and infiltration of inflammatory cells evident for acute pancreatitis in Chabro birds.

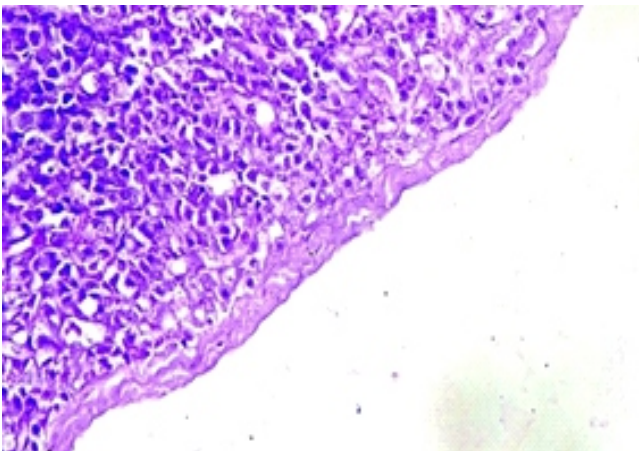


Fig.4: Photomicrograph showing fibrous thickening of pancreatic capsule without infiltration of inflammatory cells in Chabro birds. (H&E Stain; 1000X).

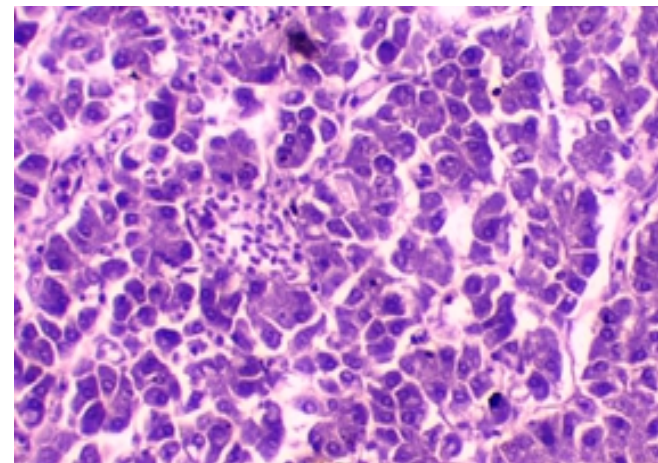


Fig.5: Photomicrograph showing acinar cells over distended with zymogen granules with nucleus situated in the basal zone and Islets of langerhan's (H&E Stain; 400X).

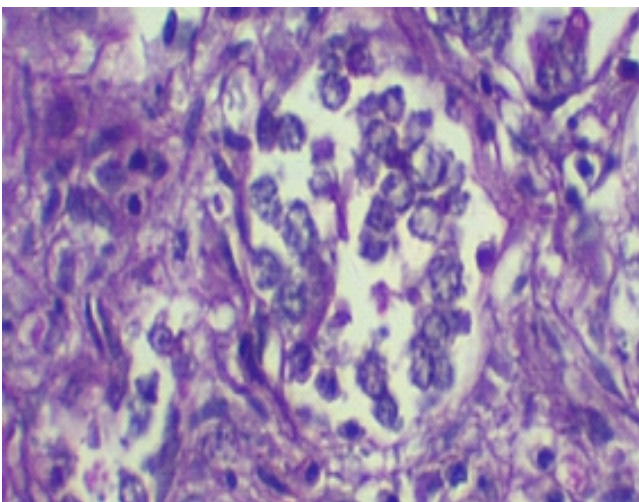


Fig.6: Photomicrograph showing Acinar cells distended with Zymogene granules. (PAS stain -400X)

membrane of acini and adjacent interstitium leading to acinar cell dissociation which is preceded by progressive depletion of zymogen granules and cell shrinkage.

CONCLUSION

It has been observed that pancreatic disorders were more common in metabolic and chronic diseases as compared to other infectious poultry diseases. The various histopathological alteration revealed that significant pancreatic pathology and their dysfunctions are an important contributory factor in chronic disease of known or idiopathic origin. Hence, supplementation of pancreatic enzymes in poultry ration in case of disease outbreak/poor growth performance are suggestive for optimal functioning of pancreas and in turn assurance of healthy poultry flock.

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