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Histopathological Changes in Small Intestine of Rotavirus Infected Poultry

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ABSTRACT

Background: Rotaviruses are important cause of acute gastroenteritis. Group A and D rotavirus are the predominant enteric viruses groups in birds. Outbreaks of rotavirus may lead to significant economic losses in poultry industry. Rotavirus infection alters the function of the small intestinal epithelium, resulting in diarrhea.

Methods: Poultry intestinal samples were collected in 10% formalin and duodenum, jejunum and ileum were processed for histopathological examination by H and E staining.

Result: Histopathological changes were noticed in all the three parts of the small intestine namely duodenum, jejunum and ileum in poultry intestinal content sample positive for AvRVD in RT-PCR. Duodenum showed necrosis, desquamation and loss of enterocytes from the villi. The jejunum showed severe disruption of villous architecture with vacuolation and separation of mucosal epithelial layer. Ileum showed a complete loss of enterocytes from the villous surface, congestion at the villous tips and infiltration of lymphocytes throughout the mucosa as well as submucosa.

Key words: Avian rotavirus, RVD, Histopathology, Intestine.

INTRODUCTION

Rotaviruses (RVs) of the family *Reoviridae* are important gastrointestinal pathogen widely distributed in humans, mammalian animals and birds (Singh *et al.,* 2015; Singh *et al.,* 2017). Rotaviruses of species A (RVA), D, F and G have been detected in birds with the predominance of RVA and RVD (Dhama *et al.,* 2015). Turkeys and chickens are maximum infected but infection in pigeons, pheasants, parrots and parakeets have been reported. Avian rotaviruses are linked to symptoms of diarrhea, reduced feed intake, growth retardation and increased mortality, even though co-infections with other pathogens seem to play a role for disease development (Jindal *et al.,* 2014). The young birds (1-2 weeks) are more susceptible for infection (Yamamoto *et al.,* 2011). RVD has also been identified as one of the causes of runting and stunting syndrome (Otto *et al.,* 2006) and RVA has been the etiology of young pigeon disease syndrome causing systemic infections with high mortality (Harzer *et al.,* 2020). Avian rotaviruses cause colossal losses to the poultry industry, as they result in decreased feed adsorption, ultimately leading to reduced weight gain (Dhama *et al.,* 2015).

Rotavirus infection alters the function of the small intestinal epithelium, resulting in diarrhea. The diarrhea is generally considered to be malabsorptive, secondary to enterocyte destruction (Kapikian *et al.* 2001). The present study was aimed to study the histopathological lesions in intestinal sample positive for rotavirus.

MATERIALS AND METHODS

Sample

The duodenum, jejunum and ileum parts of poultry samples screened for avian rotavirus were kept in 10% formalin for histopathology. The samples which tested positive for AvRVD in reverse transcriptase-polymerase chain reaction (RT-PCR) for VP6 and NSP5 genes collected during September 2017 and December 2018 from organized (Poultry Farm, DUVASU, Mathura) and unorganized poultry units / meat shops in and around Mathura region (Yaduvanshi, 2021) were used for studying the histopathological changes.

Histopathological examination

The parts duodenum, jejunum and ileum from samples (n=3) having a history of clinical diarrhoea and normal control birds were collected in 10% formalin for histopathology. After

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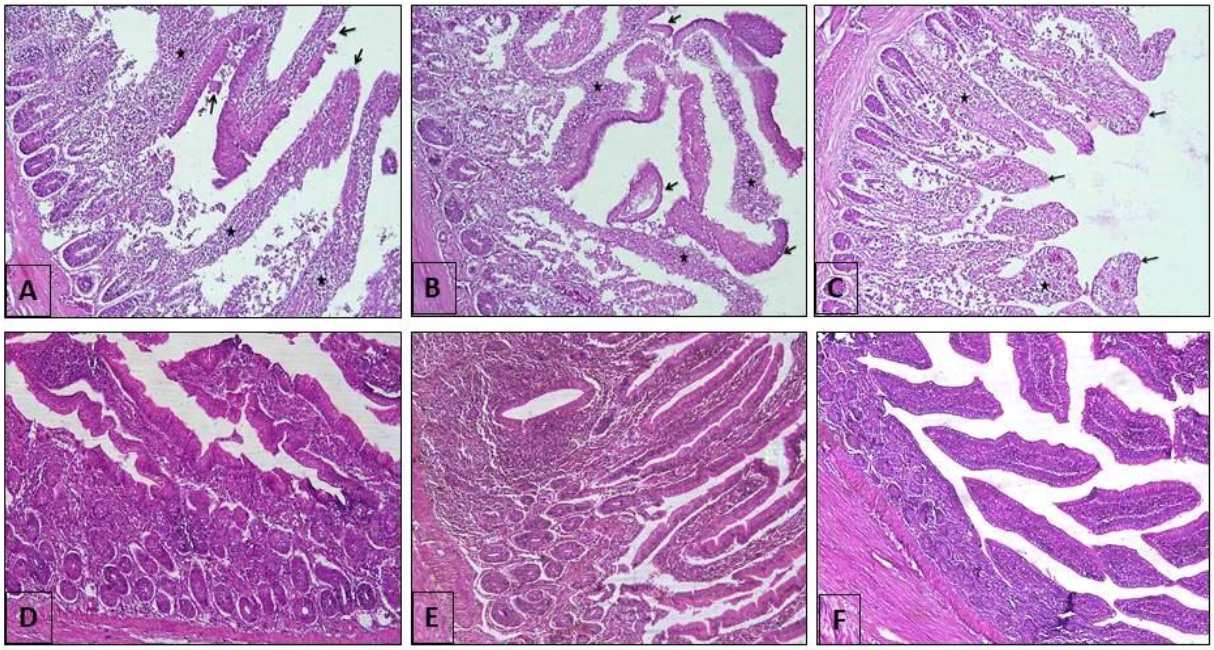

Fig 1: Histopathological changes in small intestine of poultry positive for rota virus.

A: Duodenum showing significant desquamation and loss of enterocytes (arrow) and infiltration of a large number of inflammatory cells in the lamina propria (star). **B**. Jejunum showing vacuolation and separation of enterocytes from the lamina propria (arrow). The villi are severely damaged and distorted. A significant amount of inflammatory cells (star) are seen infiltrating the lamina propria. **C**. Ileum showing a complete loss of enterocytes from the lamina propria and a large number of lymphocytic infiltrates in the lamina propria. **D. E & F.** Duodenum, jejunum and ileum of a normal healthy control of same age is given for comparison. The duodenum, jejunum and ileum show normal tissue architecture with intact villi and enterocytes. H & E × 100.

proper fixation the tissue samples were washed overnight in running tap water to remove the formalin. The tissues were then dehydrated ascending grades of alcohol *i.e.* 50%, 70%, 80%, 90%, 100%, 100%, 100% ethanol for 30 min each. Then the tissues were treated in 100% xylene two changes for 30 min each for clearing. This was followed by paraffin impregnation done by keeping the tissues in two changes of melted paraffin at 62°C for 1 hour each. After this tissue blocks were prepared and allowed to cool. The blocks were then sectioned with a microtome at 5 µm thickness. The tissue sections were collected on grease free glass slides and prepared for routine haematoxylin and eosin staining (Luna, 1968).

RESULTS AND DISCUSSION

Histopathological findings

Histopathological changes were noticed in all the three parts of the small intestine namely duodenum, jejunum and ileum (Fig 1). Duodenum showed necrosis, desquamation and loss of enterocytes from the villi. Massive infiltration of lymphocytes was also seen in the lamina propria extending deep down the crypts. The jejunum showed severe disruption of villous architecture with vacuolation and separation of mucosal epithelial layer. Infiltration of lymphocytes could also be seen in the submucosa. Ileum showed a complete loss of enterocytes from the villous surface, congestion at the villous tips and infiltration of lymphocytes throughout the mucosa as well as submucosa.

Several authors have also reported the presence of moderate-to-severe villous atrophy, fusion of villous tips, and attenuation of surface epithelium in small intestines (Otto *et al.,* 2006). Vacuolation of enterocytes, desquamation of enterocytes, shortening and fusion of the villi have also been observed in earlier studies (Salim *et al.,* 1995). Infiltration of lymphocytes and widening of lamina propria as observed in this study are common histological findings in avian rota viral infections (Kim *et al.,* 2020). In a study by Abdullazeez *et al.* (2018) in mice model histological changes in ileum were characterized by desquamation and necrosis of crypts with villous atrophy, infiltration of lymphocytes in inter villous areas and decreased population of lymphocytes in Peyer's patches and mesenteric lymph nodes. However neither gross nor histopathological changes are considered pathognomonic for the disease caused by rota viruses (McNulty, 2008). As in the present study, possibility of other etiological agents was not ruled out, the characteristic signs of enteritis found in rotavirus positive samples could not be ascertained for rotavirus infection.

CONCLUSION

This study highlights the histopathological changes in the intestinal portions of poultry that was tested positive for

Group D avian rotavirus in RT-PCR analysis. All three parts i.e. duodenum, jejunum and ileum showed the changes that have been earlier reported in rota viral infections though the changes are not considered pathognomonic.

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