Short Communication

Histological study of Broiler chicken liver infected with Marek's Disease (MDV)

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Abstract

In chicken the Marek's disease virus (MDV) is able to cause a variety of syndromes and pathological symptoms. In order to conduct the histopathology, the liver was obtained from broiler chicken in poultry estate, near Gadap, Sindh, the age of birds was 07-10 weeks. Birds had diarrhea, dropping of wings, weight loss, a grey abdomen, anorexia and keel (breastbone) was prominent and protruded out. The disease had a daily mortality rate 0.16 to 0.20 percent of the flock. On postmortem the infected liver was swollen and had lesions, a portion was removed and kept in 10% formalin for 3 days in glass vials and then send to Parasitology Section, Department of Zoology, University of Karachi. Different concentrations of ethanol were used to dehydrate tissue later embedded in paraffin wax at 52°C for 4 days, 10 µm sections were stained with haematoxylin and eosin (H & E). The prepared slides were then observed under the microscope ($\times 40$) and examined for any histological findings. It was observed that several neoplastic foci pleomorphic cells were beginning to occur and it was infiltrated with lymphomatous lesions consisting of lymphoblast and mostly elongated and small to medium sized lymphocytes. These lesions are later transferred into tumor cells and infiltrate different organs and tissues. The occurrence of MDV in 07-10 weeks old broiler chicken even after vaccination could be due to poor biosecurity conditions.

Keywords: Chicken; Histology; Liver; Marek's disease; Pakistan; Sindh

Introduction

Marek's disease is caused by an alpha-herpes virus which has the capability to induce lymphoproliferative lesions in chicken viscera and its control has been undermined by pathogen evolution. Marek's disease virus (MDV) the causative agent of Marek's cause considerable economic burden on eggs and meat

production, almost causing 1 billion USD loss per year [1].

It is also an immunosuppressive disease and causes increased susceptibility to other infections throughout the world. Marek's disease occurs in both broiler and layers in Pakistan and Pakistan is eleventh number country and produces 1163 million broiler chickens per year [2]. It has been reported from a number of countries including

Bangladesh, China and India [3-5]. Clinical signs and histopathological study is still considered a reliable aid in determining the extent of damage to liver was investigated.

Materials and Methods

The poultry farm from which chickens were received is located in poultry estate, Gadap (25.0023°N, 67.1321°E), Karachi, Sindh, Pakistan and harbors both broiler and laying chickens. The farm stock around 12,000 birds was divided into six layer flocks and four broiler flocks. The diseased broiler chickens were vaccinated with HVD subcutaneously along with IB, IBD and Newcastle intraocularly on the first day of age. Birds when autopsied were approximately 07-10 weeks old broiler chickens. The disease had a daily mortality rate 0.16-0.20% of the flock.

All the fourteen birds displayed the following clinical symptoms diarrhea, dropping of wings, weight loss, a grey abdomen, anorexia and keel was prominent and protruded out on postmortem of the birds, the liver was swollen and had lesions, a portion of liver was carefully removed from a bird with a sharp scissor and forceps and kept in 10% formalin and transferred to glass vials and then send to Parasitology section, Department of Zoology, University of Karachi. The liver tissue was dehydrated in different concentrations of ethanol and embedded in paraffin wax at 52°C for 4 days. Staining of 10 µm sections was done with haematoxylin and eosin (H & E) [6].

Results and Discussion

All the fourteen autopsied birds showed small to big lesions in the liver. In histopathological examination of liver samples for subclinical form of MD, neoplastic foci plemorphic cells were observed (Fig. 1). In some section lymphomatous lesion consisting of lymphoblasts and lymphocytes were prominent (Fig. 2), which were mostly elongated and small to medium sized (Fig. 3). The compositions of cellular tumors are generally similar between organs, but at times the involvement may vary [7]. Wen *et al.* [8] have detected presence of MDV in

nucleus of lymphoid tumor cells in the liver with localization of the Meg Protein which was subcellular in the nucleus although no MDV Meg expression in hepatic cells of the infected bird was observed. The protein is considered major factor that cause tumors [9]. The visual lesions of Marek's disease were classified by Yamamoto *et al.* [10] into three categories according to histological characteristics (i) lymphogranulomatous lesions (ii) reticulosarcoma like or lymphosarcoma like lesions (iii) Lymphoblastoid lesions. Sun and Cui [11] suggested that infection with MD leads to various illnesses that may cause death of the bird and the susceptibility to different diseases is increased. Fujimoto *et al.* [12] recorded infected liver cell proliferation which was more pronounced in the interlobular connective tissue, especially around the small blood vesicles. Furthermore they found that the lesions in tumor consisted of proliferated tumor cells which may come from lymphoreticular cell originating from the extra-capillary reticular tissue.

Boodhoo *et al.* [13] reported that vaccines are only capable of preventing neuropathy, oncogenic disease and immunosuppression but is unable to control MDV transmission or infection leading to emergence of increasing virulent pathotypes. The age of bird at the time of virus exposure has only been studied superficially [14]. Control of MD can be successfully achieved through vaccination with live attenuated vaccines, which improves genetic resistance an important component of disease but at the same time continuous evaluation of virulence and hypervirulent pathotype emergence remains a major issue for sustainable control of the disease [15].

In an earlier detail study Haq *et al.* [16] reported from Pakistan the prevalence along with risk factors of Marek's disease in commercial layer flocks in birds ranging from 13 to 30 weeks, mortality ranged from 5.40 to 11.60 percent, whereas the most important risk factor was exposure to coccidiosis, Gumboro and infectious

anaemia, howsoever in the present study the broiler chickens were infected and the daily mortality rate percentage ranged from 0.16-0.20.

Abreu *et al.* [17] reported more prevalence of disease in poultry kept in alternative systems when compared to conventional commercial establishments. The occurrence of MD in birds even after vaccination could be due to poor biosecurity condition, as the infection in free-range facilities is more widespread [18] which could explain the occurrence of Marek's disease virus in 20-25 weeks old incubated chicken. Marek's vaccine is not permitted in Iran for broiler chicken, thus

good biosecurity of broiler farms for improving the MD outbreaks along with better immune response is attained [19]. Besides vaccination proper biosecurity is necessary. Chicken are mostly infected by inhalation of severely polluted dust from poultry houses and sine virus have complex life cycle it is shed from infected birds feather follicle [20]. Chickens should be purchased from breeders or hatcheries which vaccinate their birds on first day of hatching while coop should be ventilated and properly cleaned and the most important is that infected birds must be immediately separated in order to keep the flock thriving.

Figure 1. Photomicrograph of chicken liver showing beginning of neoplastic foci pleomorphic cells (→) of chicken liver (× 200)

Figure 2. Photomicrograph showing lymphoid lesion () consisting of lymphoblasts and lymphocytes (× 400)

Figure 3. Photomicrograph showing mostly elongated and small (→) to medium size (⇒) lymphocytes in liver of chicken infected with Marek's disease virus (MDV) (× 400)

Conclusion

Marek's disease is a neoplastic and lymphoproliferate disease among chicken which is caused by MDV (Herpesviridae). In this study the flock was vaccinated on the first day of age yet occurrence of Marek's disease may be due to unsatisfactory biosecurity conditions and inhalation of contaminated dust from poultry houses in Gadap, Sindh, Pakistan. Thus, histopathology of liver was conducted to see the changes caused by MDV. The major changes observed were neoplastic foci pleomorphic cells were seen along with lymphomatous lesion consisting of lymphoblast and lymphocytes which were mostly elongated and small to medium sized.

Authors' contributions

Conceived and designed the experiments: A Khan & M Akram, Performed the experiments: N Khatoon, Contributed materials: Noor-un-Nisa & A Khan, Wrote the paper: S Waheed.

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