

Outbreak of Newcastle Disease in a Turkey Farm in Andhra Pradesh

Deepthi. B^a and Chaitanya. Y^{*b}

^aDepartment of Veterinary Microbiology, College of Veterinary Science, Garividi, Andhra Pradesh, India-521102

^bDepartment of Veterinary Clinical Complex, NTR College of Veterinary Science, Gannavaram, Andhra Pradesh, India-521102

Abstract

The present study reports incidence of Newcastle disease in turkeys housed in an integrated avian farm. Mortality recorded was more than 75% and noticed in both young and adult birds. Infected birds exhibited clinical signs including anorexia, depression, greenish diarrhea along with nervous signs few hours before death. Newcastle disease was confirmed by haemagglutination inhibition test and RT-PCR targeting partial fusion gene. All the infected birds were immediately segregated from the other birds and treated appropriately. This incidence highlights the need to vaccinate all avian species including chicken against Newcastle disease.

Keywords: Newcastle disease; Turkeys; HI; RT-PCR;

Newcastle disease (ND) is a highly infectious viral disease of avian species caused by virulent strains of Avian Paramyxovirus-1 (APMV-1). Wide variety of avian species have been reported to be susceptible to ND (Kaleta and Baldauf, 1988). Severity of the disease and the associated consequences, led to recognition of ND as OIE list A disease (Office Internationale des Epizooties, 2004). Turkeys are considered to be highly resistant to experimental infection with NDV (Tsai *et al.*, 1992; Alexander, 2000). Here we report an incidence of Newcastle disease in turkeys reared in an integrated avian farm in Tanuku, West Godavari district, Andhra Pradesh.

A total of 65 turkeys of different age groups were housed along with other avian species such as chicken, pigeons and quails during November, 2018. The clinical signs were initially noticed in five-month-old poults which included inappetence, depression, greenish diarrhea and nervous signs like neck and leg paralysis (Fig 1). Nervous signs became severe few hours before death. More than 90 percent of the birds exhibited similar clinical signs.

However, average mortality recorded in the poults and adult turkeys was 75 percent (49/65). Postmortem examination revealed focal hemorrhages and necrotic foci in the caecal tonsils and intestines, hemorrhages at the tips of proventricular glands, diffuse congestion in the viscera including kidneys, heart, lung and trachea (Fig 2). During necropsy, samples

viz., proventriculus, lung, kidneys, intestine (including contents), caecal tonsils, spleen, brain, liver and heart tissues as a pool were collected from each bird. The tissues were processed and inoculated into 9-day old embryonated chicken eggs through allantoic route. Embryos that died after 24 h of inoculation were harvested and the allantoic fluid was tested for haemagglutination. Presence of NDV was confirmed by haemagglutination inhibition (HI) test employing NDV-LaSota hyper immune serum (OIE, 2012). Reverse transcription PCR employing universal primer set targeting hyper variable region of the fusion gene i.e., fusion protein cleavage site (FPCS) (Nantha Kumar *et al.*, 2000) was also used for confirmation of NDV.

NDV was isolated from tissues collected during necropsy. Embryos dead after 24 h were harvested and observed for typical lesions which include peri occipital hemorrhages and diffuse congestion of the embryos. As a preliminary test, amnio allantoic fluid (AAF) collected from each embryo was tested by spot haemagglutination test. AAF positive by spot haemagglutination test was quantified by microtitre haemagglutination assay and the titre was found to be 1 in 2⁸. Haemagglutination activity of the sample was specifically inhibited by LaSota specific hyper immune serum in HI test. For RT-PCR, total RNA was extracted from the HA positive amnio allantoic fluid by TRIzol method. In parallel, RNA was also extracted from LaSota vaccine and allantoic fluid of uninoculated embryo are used as positive and negative controls respectively. The cDNA concentration was measured and adjusted to a minimum concentration of 100 ng and then used as template in PCR. Partial fusion

*Corresponding author: chaitanya.yalavarthi@gmail.com.

gene with an amplicon size of 356 bp was amplified as visualized in 2 per cent agarose gel containing ethidium bromide along with 100 bp DNA ladder (Fig 3).

Besides chicken, Newcastle disease was reported in different avian species such as pigeons, cormorants, psittacines, pheasants, peafowl, wild waterfowl and shorebirds (Pearson and McCann, 1975; Garcia *et al.*, 2013). Turkeys are generally considered resistant to ND infection (Alexander, 2001). In the present investigation, seventy five per cent of the affected turkey flock succumbed to infection. Several authors previously reported that mortality can easily

reach 100 per cent in viscerotropic velogenic Newcastle disease and neurotropic velogenic Newcastle disease outbreaks (Aldous and Alexander, 2001, Cattoli *et al.*, 2011 and Ganar *et al.*, 2014). Balachandran *et al.* (2014) reported 3-7 per cent drop in egg production in ND affected flocks. Integrated or mixed poultry farming is a new trend resulting in good returns to the farmers. Nevertheless, there is every chance for spill over of pathogens among different avian species. Furthermore, vaccination against ND is focused and limited only to the commercial poultry sector which makes other avian species vulnerable.



Fig. 1: Turkey poult exhibiting leg and neck paralysis along with greenish diarrhea

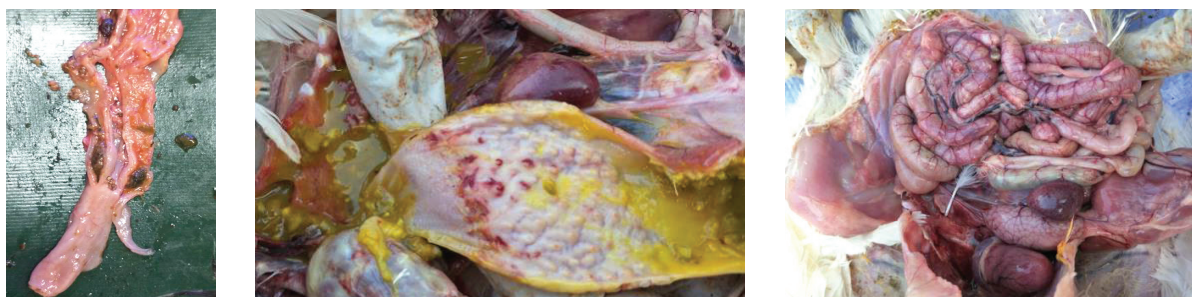


Fig. 2: Postmortem lesions- Necrotic foci in the caecal tonsils, haemorrhages on the proventricular glands and visceral congestion.

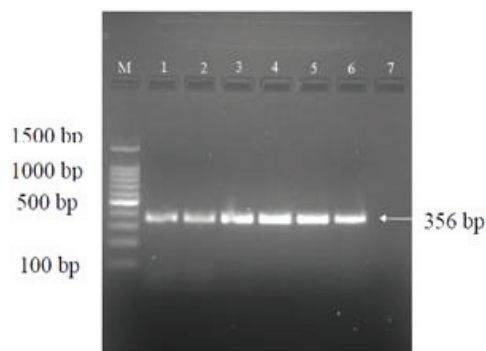


Fig. 3: RT-PCR for partial fusion gene. M- 100 bp marker; Lanes 1-5: Positive samples; Lane 6: Positive control; Lane 7: Negative control

Acknowledgement

The authors are thankful to Sri Venkateswara Veterinary University, Tirupati for providing financial support to carry out the present study.

References

- Alders, R.G. 2014. Making Newcastle disease vaccines available at village level. *Vet. Rec.* **174** (20), 502-503.
- Aldous, E.W. and Alexander, D.J. 2001. Detection and differentiation of Newcastle disease virus (avian paramyxovirus type 1). *Avian Pathol.* **30**(2): 117-128.
- Alexander, D.J. 2000. Newcastle disease and other avian paramyxoviruses. *Revue Scientifique et Technique-Office International des Epizooties*, **19**(2): 443-462.
- Balachandran, P., Srinivasan, P., Sivaseelan, S., Balasubramaniam, G.A. and Murthy, T.G.K. 2014. Isolation and characterization of Newcastle disease virus from vaccinated commercial layer chicken. *Vet. World.* **7**(7):457-462.
- Cattoli, G., Fusaro, A., Monne, I., Molia, S., Le Menach, A., Maregeya, B., Nchare, A., Bangana, I., Maina, A.G., Koffi, J.N., Thiam, H., Bezeid, O.E., Salviato, A., Nisi, R., Terregino, C. and Capua, I. 2010. Emergence of a new genetic lineage of Newcastle disease virus in West and Central Africa – Implications for diagnosis and control. *Vet. Microbiol.* **142** (3-4): 168–176.
- Dimitrov, K.M., Abolnik, C., Afonso, CL., Albina, E., Bahl, J., Berg, M., Briand, F.X., Brown, I.H., Choi, K.S., Chvala, I. and Diel, D.G. 2019. Updated unified phylogenetic classification system and revised nomenclature for Newcastle disease virus. *Infect. Genet. Evol.* **74**:103917.
- Ganar, K., Das, M., Sinha, S. and Kumar, S. 2014. Newcastle disease virus: current status and our understanding. *Virus res.* **184**: 71-81.
- Garcia, C.S., Lopez, R.N., Morales, R., Olvera, M.A., Marquez, M.A., Merino, R., Miller, P.J. and Afonso, C.L. 2013. Molecular epidemiology of Newcastle disease in Mexico and the potential spillover of viruses from poultry into wild bird species. *Appl. Environ. Microbiol.* **79** (16): 4985–4992.
- Kaleta, E.F and Baldauf, C. 1988. Newcastle disease in free-living and pet birds. In: Newcastle disease , Springer, Boston, MA. pp 197-246.
- Miller, P. J. and Koch, G. 2013. Newcastle disease. *Dis. Poult.* **13**:89–138.
- Nantha kumar, T., Kataria, R.S., Tiwari, A.K., Butchaiah, G. and Kataria J.M. 2000. Pathotyping of Newcastle disease viruses by RT-PCR and restriction enzyme analysis. *Vet. Res. Commun.* **24**(4): 275-286.
- OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. 2004. Newcastle disease. Chapter 2.1.15. <http://www.oie.int>.
- OIE Manual of diagnostic tests and vaccines for terrestrial animals: mammals, birds and bees, 2012. Newcastle disease. Biological Standards Commission. World Organisation for Animal Health, Paris, France. 555–574.
- Pearson, G. L. and Mccann, M. K. 1975. The role of indigenous wild, semi domestic, and exotic birds in the epizootiology of velogenic viscerotropic Newcastle disease in southern California, 1972-1973. *J. Am. Vet. Med.* **167**(7): 610-614.
- Taxonomy IV. Release. 2019. International committee on taxonomy of viruses. v/ msw054.
- Tsai, H.J., Saif, Y.M., Nestor, K.E., Emmerson, D.A. and Patterson, R.A. 1992. Genetic variation in resistance of turkeys to experimental infection with Newcastle disease virus. *Avian diseases*, pp.561-565.