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A case report on concurrent infection of Newcastle disease and Infectious bronchitis in pigeon

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Abstract
A six month -old pigeon with clinical signs of diarrhoea and torticollis was presented to the Department of Veterinary Microbiology, College of Veterinary and Animal Sciences, Mannuthy for disease investigation. The bird was sacrificed and samples from lungs, trachea, air sacs, liver, spleen, heart blood, intestine and brain were collected aseptically for isolation of bacteria and fungi. Nucleic acid was extracted from the tissue samples collected and were subjected to polymerase chain reaction (PCR) assays for the detection of Newcastle disease virus, Infectious Bronchitis virus, Infectious Laryngotracheitis virus and *Mycoplasma gallisepticum.* No organism of pathogenic significance could be isolated by bacterial and fungal culture. On PCR, amplicons could be detected at 236 bp and 386 bp, respectively for Newcastle disease and Infectious bronchitis.

Keywords: concurrent, newcastle, infectious bronchitis, pigeon

1. Introduction
Pigeons has been associated with humans from the ancient times and they live side by side with humans as a source of food, hobby and for experimental purposes. It has long productive life and short reproductive cycle. Besides these, pigeons are susceptible to various infectious diseases. Among them, Newcastle disease (ND) and infectious bronchitis (IB) play significant role in causing mortality and decrease in production performance of the birds. In addition, they cause huge economic losses to the owners. Newcastle disease virus (NDV) infection has been reported in pigeons and is primarily associated with neurological signs. The NDV isolates obtained from diseased pigeons are termed as pigeon paramyxovirus-1. Infectious bronchitis is an acute and highly contagious viral disease that affects the respiratory tract of birds of all ages. The etiological agent of IB is Infectious Bronchitis virus (IBV), placed in Group 3 of genus *Coronavirus* of the family *Coronaviridae*.

2. Materials and Methods
The owner reported that there was huge mortality in a flock of pigeons at Muthuvara, Thrissur district of Kerala and the birds were showing symptoms of diarrhoea and neurological disorders such as torticollis before death. The ailing bird from the flock was brought to the Department of Veterinary Microbiology, College of Veterinary and Animal Sciences, Mannuthy for disease investigation. The birds were sacrificed and the samples (lungs, trachea, air sacs, liver, spleen, heart blood, intestine, brain) were collected aseptically for bacterial and fungal culture and nucleic acid extraction. For bacteriological examination, samples from, lung, trachea, air sacs, liver, spleen, heart blood and intestine were streaked on to bovine blood agar and brain heart infusion agar. Liver and intestine were also streaked on to MacConkey agar and intestine was inoculated in selenite broth and incubated at 37˚C for 24 h. For fungal culture, samples in duplicate were streaked onto Sabouraud's dextrose agar and incubated at 37℃ and at room temperature. For virological examination, tissue samples were collected in a sterile RNAse free vial with RNA later. The tissue samples were homogenised and subjected to RNA and DNA extraction. The RNA was extracted using Trizol method. The extracted RNA was converted to cDNA using commercial cDNA synthesis kit. The DNA was extracted using Himedia HiPurA™ Multi- Sample DNA purification kit. The converted cDNA and the extracted DNA were subjected to polymerase chain reaction (PCR) assays for the detection of Newcastle disease virus, Infectious Bronchitis virus, Infectious Laryngotracheitis virus and *Mycoplasma gallisepticum.*

3. Results and Discussion

On Blood agar, Brain heart infusion agar and on MacConkey agar, no bacteria could be isolated even after 48 h of incubation. No growth could be detected on SDA at 37˚C and at room temperature even after seven days of incubation. The PCR assays for Infectious laryngotracheitis and *Mycoplasma gallisepticum* did not reveal any positive results. Positive amplicons could be detected at 236 bp and 386 bp, respectively for ND and IB (Fig. 1). This is in accordance with Kho *et al.* (2000) [3], Akin *et al*. (2001) [1] and Zaher and Girh (2014) [4] who reported concurrent infections of ND and IB in chickens. Hadipour *et al.* (2011) [2] also reported concurrent infection of Avian Influenza, ND and IB in commercial broiler farms. The present case study reports the concurrent infection of ND and IB in pigeons in Kerala. The study highlights that there is an urgent need for the implementation of routine laboratory surveillance for viral pathogens not only for chickens but also for other poultry species, as the transmission can easily occurs between the poultry species. The report also stresses the need for the development of strategies to improve the quality of vaccine and vaccination of the poultry flocks in Kerala to safe guard the poultry industry.

4. Acknowledgement

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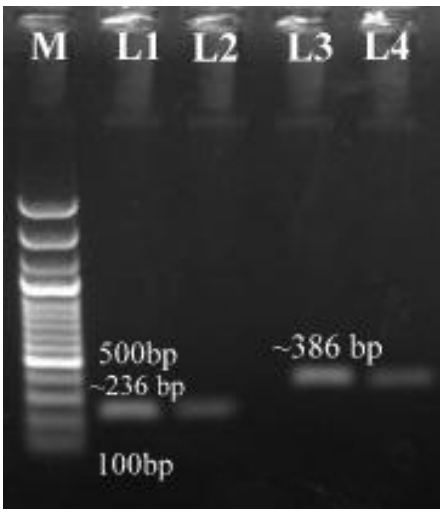

Fig 1: Amplicons for ND and IB

Lane M: Molecular ladder 100bp (Thermoscientific, USA)
L1: Positive control for ND
L2: Positive sample for ND
L3: Positive control for IB
L4: Positive sample for IB

5. References

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