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ISOLATION OF NEWCASTLE DISEASE VIRUS FROM TEALS *(Anas crecca)* IN IRAN

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Abstract: Eight of 30 teals *(Anas crecca)* died several days following capture and Newcastle Disease Virus (NDV) was isolated from all eight. Brains from the dead birds were homogenized and inoculated into chicken embryos. The allantoic fluid from the embryos was inoculated into 10 domestic chickens susceptible to NDV and 10 chickens immunized against NDV. Eight of 10 (80%) susceptible chickens died, while the immunized chickens remained healthy. Anti-NDV serum showed complete homology against NDV and the eight isolates.

INTRODUCTION

Waterfowl generally are not refractory to Newcastle disease virus (NDV). Isolation of NDV have been made from ducks *(Anas acuta, Anas cyanoptera, Anas crecca carolinensis*[6] *Anas discors* and *Anas carolinensis,*[8] *Anas platyrhynchos,* and *Anas streperal)*[10] and geese *(Branta canadensis).*[5,8] Antibody against NDV has been demonstrated in ducks (unidentified),[6] *(Anas acuta tzitzihoa)*[7] and Canada geese *(Branta canadensis interior).*[2]

This report describes the isolation of NDV from free-ranging teal *(Anas crecca).* In December, 1976, eight of 30 teals died shortly after capture in the Caspian sea coast region. The teals had been caged in a house with no possibility of contact with other birds at the time.

MATERIALS AND METHODS

Brains were removed and were homogenized in phosphate buffered saline (PBS) containing antibiotics for viral isolation. Homogenized samples were centrifuged at 1500 × g for 20 min and the supernatant of each sample inoculated into the allantoic cavity of three 9-day-old chicken embryos. Along with this procedure five eggs also were inoculated with PBS as controls. The eggs were incubated at 39 C and examined daily for four days post-inoculation or until death of the embryo. Following death of the embryos (2 to 4 days post-inoculation), the allantoic fluid was removed and stored for viral identification. There was no work on NDV during the time these specimens were processed.

Immune serum was prepared by inoculating a rooster intramuscularly with a known strain of ND (Zabol strain) four times at weekly intervals. Serum was collected two weeks after last inoculation and used in the serum neutralization (SN) test. The standard hemagglutination (HA), hemagglutination-inhibition (HI) and SN tests were used to identify the virus. The SN test was conducted using chicken fibroblast cells. Chicken erythrocytes were used in the HA and HI tests.

Ten 20-day-old domestic chickens susceptible to NDV were inoculated with allantoic fluid obtained from the chicken embryos. Another group of ten 20-day-old chickens vaccinated against NDV also were inoculated with the fluid.

The surviving teals were not available for serologic tests.

RESULTS

Routine post-mortem examination revealed all eight birds were in good physical condition. The only obvious

changes seen were general darkening of the breast and leg muscles and petechial hemorrhages on the heart and on the abdominal fat deposits.

All embryonated eggs inoculated with the homogenate from the brains died between 48 and 96 h post-inoculation. Allantoic fluid from the eight samples showed hemagglutination titers ranging from 1/64 to 1/512. Anti-NDV serum diluted to a titer of 1/128 inhibited eight hemagglutination units of all isolates. None of the control embryonated egg died during the experiment.

Anti-NDV serum showed complete homology between NDV and the eight isolates from teal. One hundred $TCID_{50}$ of all isolates were neutralized by a 1/64 dilution of NDV antiserum.

Eight of the 10 (80%) susceptible chickens exposed to one of the isolates developed clinical signs of respiratory distress followed by clinical signs of central nervous system involvement 4 to 6 days post-inoculation, and died 3 to 5 days later. The two remaining chickens in the susceptible group, and all 10 chickens in the immunized group remained healthy for the duration of the experiment (20 days).

DISCUSSION

The results of our report and others[1,3,6,8,10] indicate that waterfowl appear to be susceptible to NDV, and under some conditions the disease has occurred and produced a high rate of mortality.

Although the exact origin of the ND infection in this report is not known, the high spread of NDV in domestic fowl flocks in Iran, some of which have poor management, suggest that spread from domestic poultry must occur. The susceptibility of teals to the disease may be of considerable importance because as migratory species they are potentially capable of disseminating the virus over long distances.

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