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CHARACTERIZATION OF A NEWCASTLE DISEASE VIRUS ISOLATED FROM A PARROT *(Psittacus erythracus)* IN NIGERIA

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Abstract: The characteristics of a Newcastle disease virus isolated from a parrot *(Psittacus erythracus)* in Nigeria were examined using standard laboratory tests. Minimum lethal dose in embryos was 10^{-10}, mean death time 44.8 h. The intracerebral and intravenous pathologic indices were 1.65 and 2.42, respectively. The virus was resistant at pH 3 and pH 7.2 and the hemagglutinin was thermostable at 56 C for 120 min. Of 10 mammalian species of erythrocytes examined, those of equine and rat were not agglutinated. The isolate was typed as a velogenic viscerotropic Newcastle disease virus.

INTRODUCTION

Newcastle disease has been present continuously in Nigeria since 1951, occasionally in epidemic proportions. A seasonal variation in the incidence of the disease in the country is apparent, more outbreaks occurring in the dry season (October-March) than in the wet season (April-September).

Over a 14-year period (1965-1978), a total of 284 suspected Newcastle disease virus (NDV) outbreaks were examined at the National Veterinary Research Institute, of which 140 (49%) were confirmed, all in domestic poultry. Thirteen NDV isolates obtained from these outbreaks have been characterised and all were found to be the velogenic strain.[3] In 1979, the laboratory investigated 31 outbreaks in domestic poultry and 21 (67%) were positive. In January of the same year, NDV was isolated from a captive African grey parrot *(Psittacus erythracus)* received for diagnostic examination from a local Wildlife Reserve.

This is the first isolation of Newcastle disease virus from a parrot in Nigeria and this paper describes certain characteristics of the isolate.

MATERIALS AND METHODS

Virus identification. The isolate was obtained by inoculating samples of lungs and proventriculus from the parrot into the allantoic cavity of 10-day-old chicken embryos and was identified as NDV by the hemagglutination-inhibition test using a standard positive Newcastle disease serum and a normal serum.

Typing of the virus. Details of the techniques have been described.[1,2] Briefly the tests were as below.

Mean death time (MDT) in embryos. Two titrations were made, in each of which the virus was inoculated into 5 embryos per dilution (10^{-1}, 10^{-7}, 10^{-8}, 10^{-9}, 10^{-10}, 10^{-11}, 10^{-12}). Observations were made at 8 hour intervals for embryo death and the presence of virus in the allantoic fluid. The highest dilution of the virus killing all embryos was noted.

Intracerebral pathogenicity index (ICPI). Ten one-day-old chicks each were inoculated with 0.05 ml of a 10^{-1} dilution of the virus. These were observed for 8 days and the ICPI calculated by standard methods.[1]

Intravenous pathogenicity index (IVPI). A 10^{-1} dilution of the virus was

inoculated in 0.1 ml amounts into 10 six-week-old chicks and the IVPI was determined following observation for 10 days.

Heat stability of the hemagglutinin. Vials of the neat and diluted virus (10^{-1} to 10^{-5}) were submerged in a water-bath at 56 C, removed at intervals of 15, 30, 60, 120 min and examined for hemagglutinin activity using erythrocytes from chicken, cattle and goat.

Stability of virus infectivity at pH 3 and pH 7.2. Following exposure at these pH solutions for 4 h, the virus was tested for infectivity in developing chicken embryos.

Hemagglutination of mammalian erythrocytes. A standard hemagglutination test was carried out with the virus using varying concentrations of erythrocytes (0.5%, 0.75%, 1%) obtained from human, cattle, goat, sheep, horse, dog, guinea-pig, mice, rabbit and rat.

RESULTS

The virus had a mean death time of 44.8 h, its minimum lethal dose being 10^{-10}. Virus was recovered from each allantoic fluid collected from the infected embryos. The ICPI was 1.65 and the IVPI 2.42. In the IVPI test, all virus-infected chicks died between three and four days and the chicks showed diffuse hemorrhages along the intestinal tract. Controls were normal and no virus was obtained at the end of each experiment.

In the heat stability test, hemagglutinin was still detectable in the undiluted virus at 120 min in all three types of erythrocytes used. Virus hemagglutinin was also demonstrated in 10^{-1} and 10^{-2} dilutions at 120 min only in chicken erythrocytes and in 10^{-1} dilution at 30 min using caprine erythrocytes. Virus agglutinin was consistently undetectable at higher dilutions (10^{-3} to 10^{-5}) in all erythrocytes. Bovine blood showed negative hemagglutinin activity in all of the five serial virus dilutions.

All embryos infected with virus following exposure to pH 3 and pH 7.2 were killed between 48 and 72 h, the LD_{50} titer being 10^5 per 0.1 ml.

The virus did not agglutinate erythrocytes from the horse and the rat at any concentration. It agglutinated bovine erythrocytes only at 0.5% concentration. Human and canine erythrocytes were negative, respectively, at 0.5 and 1% dilutions. Mouse blood was negative at 1% dilution.

DISCUSSION

Four pathotypes of Newcastle disease virus currently are recognized.[3] These are differentiated in the laboratory by standard test procedures, namely the mean death time and the intracerebral and intravenous pathogenicity tests.[1] The velogenic pathotype has a mean death time of less than 60 h and a minimum index of 1 and 0.5 with respect to intracerebral and intravenous pathogenicity.[3,4,5] The results of these tests show that the isolate examined is a velogenic strain of Newcastle disease virus with a proclivity for the visceral tissue. The virus has a high rate of growth and infectivity, as indicated by the result of the test for the mean death time.

The virus agglutinin is relatively thermostable although this appears to be largely a function of concentration. The chicken erythrocyte appears to be the most sensitive indicator of hemagglutinin activity following thermal exposure. The ability of the virus to resist low and high pH suggests that it is capable of prolonged survival and infectivity under local field conditions. In the hemagglutination test with mammalian erythrocytes, the behavior of the virus in bovine and equine blood is reminiscent of velogenic strains previously isolated from poultry in Nigeria.[5]

The apparent resistance of bovine, canine and human erythrocytes to

agglutination at higher and lower concentrations is difficult to explain, but possibly was associated with the rate of virus elution. The inability of the virus to agglutinate rat erythrocytes may be an attribute of other velogenic isolates in the country; however, if not this may be a valuable asset to differentiate strains of local isolates.

Newcastle disease has been diagnosed serologically in falcons in Kano State of Nigeria,[6] suggesting a role by wild birds in the epidemiology of the disease in Nigeria. Isolation of the virus from a parrot strongly indicates that virus is shed by serologically positive wild birds, including parrots. Thus, wild birds could complicate control programs against Newcastle disease in Nigeria, especially as vaccination of domestic poultry will not eliminate the danger of the virus circulating in free-flying species.

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