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Newcastle Disease in Village Chickens in Sudan: Survey of Disease Incidence and Isolation of the Causative Virus

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Abstract: The present study described the epidemiology of Newcastle disease (ND) in village chickens in Sudan. The study was carried out in the framework of the project; *Improving Family Poultry Production in Africa*. Five and three villages in Khartoum (zone 1) and Gedarif (zone2) provinces respectively were selected for the study. Farmers interview, virus isolation and hemagglutination (HA) and hemagglutination inhibition (HI) tests, to identify the isolated viruses, were employed. The biological characteristics of each virus isolate were also determined. Two out of 20 households (10%) in zone 1 reported occurrence of ND during the study year compared to eleven out of 12 households (91.5%) in zone 2. The disease caused a mean mortality rate of 66% and 69% in zone 1 anu 2 respectively. All age groups were found affected and the mortality rate was 70% in chicks, 98% in growers and 62% in adults. Two isolates of NDV were obtained in embryonated eggs following their confirmation by HA and HI using a reference NDV serum. The isolates were designated as GD.S.1 and GD. Gh.1. The isolates showed similarity in that they kill embryos rapidly in mean death time test, produced visceral lesions in 8-week-old chicks and had a high Intracerebral pathogenicity index (ICPI). Accordingly, the isolated viruses were grouped as velogenic viscerotropic NDV (VVNDV) pathotype.

Key words: Newcastle, chicken, disease, incidence, isolation, causative virus

Introduction

Throughout Africa, village chickens are the chief source of animal protein in rural areas (Musiime, 1992; Alemu, 1995; Tadlle and Ogle, 1996). In addition, they were reported to have a great social importance (Mukiibi, 1992). Among a population of 45.3 millions of chicken in Sudan, the conventional sector contains around 30 millions (66.2%) from which the annual meat and egg production is about 20.1 million birds and 900 million eggs respectively (Sulieman, 1996). Despite this significance, little information is available in the literature on the disease incidence among these chickens. Among the diseases of village chickens, ND was ranked as the most important disease in many parts of the world (Spradbrow, 1993).

There was no detailed study on the epidemiology of ND in village chickens in Sudan elucidating the exact effect of the disease on production .The present study is the first detailed report on the situation of ND in village poultry in the Sudan. The study is particularly, aiming at gaining more data on the effect of the disease in terms of morbidity and mortality and to isolate and characterize the ND virus (NDV) circulating in village chickens.

Materials and Methods

Study areas: Khartoum and Gedarif were the two provinces selected for the study and designated zone 1 and zone 2 respectively. Five and three villages were selected in zone1 and zone 2 respectively. Four to six households were selected in each village (numbered 1-6). The selection of Khartoum province (Centre of Sudan) and Gedarif province (Eastern Sudan) was made to represent two ecologically different environments, while the villages and households were selected based on the advice of the local Veterinary authorities taking into consideration the number of chickens per household and previous co-operation with the veterinary authorities.

Data Collection: A survey form supplied by the joint FAO/IAEA division of Animal health was used for collection of data. This form was developed to standardise the collection of production and health data on village poultry. The study areas were visited in the dry and wet seasons. During each field visit the person responsible for chicken production in each household was interviewed about the occurrence of the disease during the study year (June 1998- May 1999) and data were entered in the survey form.

Specimen Collection: Cases suspected as ND were brought to the Virology Laboratory (Faculty of Veterinary Medicine, University of Khartoum). Post-mortem examination was carried out. Spleen, lung and brain tissues were collected aseptically into sterile bijou bottles and stored at -20°C till used.

Virus Isolation and Identification: Collected tissues were homogenized and centrifuged at 1000 rpm for 10 minutes.

The supernatant was inoculated into the allantoic cavity of 9-11 day-old, embryonated chicken eggs. The allantoic fluids (AF) were collected for virus harvest. Obtained isolates were identified as (NDV) by the hemagglutination (HA) and hemagglutination inhibition (HI) tests.

Virus Characterization: The isolated NDV were characterized by determining the following parameters:

A. Chick embryo lethal dose 50% end point (CELD50%): This was essentially performed as described by Reed and Muench (1938).

B. Chick embryo mean death time of the minimum lethal dose (MDT/MLD): A number of 10, ten -day-old embryonated eggs were inoculated via the allantoic cavity with the highest dilution at which all inoculated embryos died as determined by CELD 50%. Five embryos were inoculated in the morning (8:00am) and five eggs in the afternoon (16:00pm). The inoculated eggs were inoculated and candled twice a day at intervals of 8,16 hours, death time was recorded in hours for each embryonated egg and the MDT/MLD was calculated as described by Hanson (1980) using the following formula:

$$MDT = \frac{(\text{No. dead at x hour})x(\text{Xhour}) + (\text{No. dead at y hour})\ x(\text{Yhour}) \text{ ect.}}{\text{Total number dead}}$$

C. Intracerebral Pathogenecity Index (ICPI): Four this purpose, sixteen, one- day -old chicks (divided into two groups of 8 chicks each) were used. Each group of chicks was inoculated with 0.1ml of undiluted AF of each field isolate intracerebrally in caudal part of the cranium using one ml disposable syringe fitted with 26 -gauge needle. Chicks were housed in isolators and observed daily for 8 days. The survival index, based on time of death, was calculated by categorizing each chick as normal (0), diseased (1) and dead (2). Calculation of the ICPI was done according *to the procedure of Hanson (1980).*

D. Pathogenicity of NDV isolates in 8- week– old chicks: For this purpose, a total of 8- week-old chickens were used. Undiluted freshly harvested AF of each field isolate was used to inoculate four chickens by swabbing onto the conjunctiva and the cloaca. Death time and post- mortem lesions were recorded. Identification of the different strains was done according to the procedure of Hanson (1980).

Statistics: The statistical significance of differences between groups of data was determined using the two-tailed Student's unpaired t-test.

Results

The incidence and mortality rates due to ND in village chickens in both zones of study are demonstrated in Table 1.Only two out of 20 (10%) household in zone 1 reported incidence of ND during the study period whereas 11 out of 12 household (91.5%) reported incidence of the disease in zone in the same period. The mean mortality rate in zone 1 was 66% and ranged between 55-77%, while in zone 2 was 69% and ranged between 30-96%. The mortality rates due to ND among different age groups of village chickens in both zones of study are shown in Table 2. The mortality rates due to ND were observed to be 70%, 97.6% and 62% for the chicks, growers and adult birds respectively.

Two NDV isolates were obtained from zone 2 (Gedarif province). These isolates hemagglutinated chicken red blood cells with hemagglutination (HA) titre of 6 $\log_2$ (for the first) and 7 $\log_2$ (for the second) and their hemagglutinability

Table1: Incidence and mortality rate due to Newcastle disease in village chickens in zone 1 and 2.

Zone	Village	Mortality (%)	Mean mortality rate (%)
1	Hassanya	67.50*	
	Ezerab	NR	
	Abu Halima	NR	67.5
	El Gaili	NR	
	A Dabba	NR	
2	Abbayo	75.13	
	Elsofi	73.75	72.5
	Ghibasha	68.75	

* Mortality rate (%) = No of dead birds/ total number of birds x 100.
NR = No reports of mortality due to ND in these villages.

Table 2:Mortality rates due to ND among different age groups of village chickens in zone 1 and zone 2

Zone	Village	Chicks	Growers	Adults
1	Hassanya	50*	-	28
	Ezerab	-	-	-
	Abu Halima	-	-	-
	El Gaili	-	-	-
	AL Dabba	-	-	-
2	Abbayo	100	100	60.3
	Elsofi	45	63.3	60
	Ghibasha	80.3	99.2	43

*Mortality rate (%) = No of dead birds/ total number of birds x 100

Table 3 : Biological properties of the NDV isolates obtained from village chickens in Sudan

Isolate	CELD50%	MDT/MLD per hr.	ICPI	Pathogenicity to 8-week- old chickens
GD.S- 1	10-8.3	47.2	1.78	All infected birds showed diarrhea, 70 %of birds showed paralysis, post-mortem examination revealed haemorrhages in the proventriculus, all birds died 4-5days P.I
GD.Gh- 1	10-8.6	47	1.66	All infected birds showed diarrhea,50% of birds showed paralysis, post-mortem examination revealed haemorrhages in the proventriculus, all birds died 3- 4 days P.I

CELD50% = Chick embryo lethal dose 50% end point
MDT/ MLD = Mean death time of the minimum lethal dose
ICPI = Intracerebral pathogenicity index
P.i = Post inoculation

was inhibited by a known NDV antiserum. The isolates are designated as GD.S.1 and GD.G.1. The biological properties of these isolates are summarized in Table 3.

Discussion

Search of the literature revealed very few reports on ND in village poultry in Sudan. Khogli (1971) illustrated the picture of the disease in indigenous stocks of chicks compared to foreign breed chicken. In that study, he noticed that the lesions were always severe in the local chickens and attributed this to the little control measures and vaccination programs that the local chickens received compared to those paid to the foreign breeds of chickens. The present study was carried out in two ecologically different zones in Sudan in an attempt to determine the epidemiology of ND in village chickens. The results obtained indicated that the incidence of the disease in zone 2 was significantly higher ($p < 0.05$) as compared to zone 1. This again attributed to little disease control attention made and to the high density of chicken population in zone 2 as compared with that of zone 1 which enhance rapid spread of the virus among chickens. Similar findings and observations were previously described regarding NDV spread (Khalafalla *et al.*, 2000 and Spradbrow, 1993).

Although all age groups of chickens were affected, it seemed that growers are the most susceptible age group to ND infection in village chickens. This may be explained in the way that baby chicks probably had maternal antibodies to NDV and adult chickens, with mature immune system, may be exposed to NDV at previous times (Alexander, 1991). The high mortality rates due to ND in village chickens in Sudan reported in this communication is comparable to reports from other African countries that documented earlier by (Shane, 1984; Sharma *et al.*, 1988 and Musiime, 1992).

Two NDV isolates were obtained in the present study from outbreak of ND involving indigenous chickens in two different villages in zone 2. The first isolate, designed as GD.S-1 (GD for Gedarif and S for Soufi village), was found to affect the 3-days-old chickens causing sudden death and mortality of 100% while the second isolate, designed GD.Gh-S (GD for Gedarif and Gh for Ghibaisha village), affected grower and adult chickens causing a mortality rate of 62%. These isolates showed almost closely related biological behavior and pathogenic properties when studied and consequently grouped as velogenic viscerotropic NDV (VVNDV) pathotype. The pathogenic properties of these isolates were similar to those described earlier for other Sudanese ND isolates obtained from exogenous breeds in commercial farms (Eisa, 1979; Ballouh *et al.*, 1983; khalafalla *et al.*, 1992 and Haroun *et al.*, 1992). It can, therefore, be postulated that indigenous chicken might be the source of the VVNDV, which caused outbreaks in

commercial farms because of negligence of village chickens in vaccination campaigns.
Based on that data, vaccination programs for the village chickens against ND is recommended as the perpetuation of the virus in village poultry poses a potential hazard of the disease to modern poultry sector in Sudan and elsewhere.

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