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### اليمام كعامل فـيـروس مرض النيوكاسـل

صلاح موسى ، عوض ابراهيم ، مصطفى شحاته

أمكن اكتشاف الأجسام المضادة المانعة للتلازن و/ أو الأجسام  
الترسبية لفـيـروس مرض النيوكاسـل فى ١٤ حالة من ٢٠٠ حالة  
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أمكن عزل عـتـرتين لفـيـروس النيوكاسـل من مسحات من المجمع  
ومن الأحشاء الداخلية لهذا اليمام ، هذا وقد أمكن  
توصيف هذه العـتـرات على أنها عـتـرات شـد يـد  
الضـرـاء من فيروس النيوكاسـل .

DOVES AS CARRIERS TO NEWCASTLE DISEASE VIRUS  
(WITH 3 TABLES)

BY  
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SUMMARY

Haemagglutinating- inhibiting and/or precipitating antibodies to Newcastle disease virus were detected in 41 out of 200 apparently healthy doves caught at different localities around Assiut.

Two Newcastle disease virus isolates were recovered from the droppings and internal organs of these doves. They were characterized as virulent strains of Newcastle disease virus.

INTRODUCTION

Newcastle disease (ND) has a wide host range including wild and free- living birds, which play an important role in the transmission of the virus to domestic fowls. (BAIDETTE, 1950; BRANDLY, 1947 & 1950; GALLESPIE ET AL., 1950; GUSTAFSON and MOSES, 1953; HANSON and SINHA, 1952).

KRANEVELD and MANSJOER (1950) and REUSS (1961) found that infection of spotted doves per os resulted in the excretion of ND virus for a period of at least 3 days in a sufficient quantity to infect baby chicks placed in contact.

MAGID ET AL. (1965) concluded that doves were refractory to oral infection with ND virus, but the haemagglutination-inhibition (HI) titre was slightly raised.

The present work was designed to throw light on the situation of the disease in doves and their role in disseminating the virus.

MATERIALS AND METHODS

Doves:

200 apparently healthy adult doves (*Streptopelia senegalensis aegyptiacus*) were caught from different areas around Assiut. They were wing banded and observed closely for 2 weeks for any abnormal signs. At the end of the observation period, blood samples and cloacal swabs were collected and finally the birds were sacrificed and subjected to bacteriological and virological examinations.

Chicken embryos:

9 to 11-day-old embryonated eggs were kindly supplied by Beni-Mur governmental farm and used for virus isolation.

Chickens:

- a- One-day-old Fayoumi chicks from a private poultry farm, free from maternal antibodies to NDV were used for intracerebral inoculation.
- b- Six-week-old susceptible chickens were supplied by Beni-Mur governmental farm as one-day-old and reared in strict isolation without vaccination until the suitable age. They were used for intramuscular inoculation.

Pigeons:

Squabs were obtained from the local market and used for intracerebral inoculation. They were checked for HI antibodies before being inoculated.

Reference ND virus:

The komarov strain was used in this study as reference virus. It was passaged in embryonated chicken eggs and titered  $10^{8.5}$  EID<sub>50</sub>/ml.

Detection of antibodies to ND virus:

Serum samples were tested for HI antibodies by the standard procedure with the microtechnique after ALLAN and GOUGH (1974). Four HA units of the komarov strain were used in the test. Sera were also tested for specific precipitating antibodies using the microtechnique of the agar-gel precipitin test (AGPT) after NANSI (1958). The antigen used was supernatant of homogenized chorioallantoic membranes harvested from chicken embryos that were infected with the Komarov strain.

Isolation and identification of ND virus:

Individual swabs from the cloaca, as well as samples from the internal organs (liver, spleen, kidneys, lungs and brain) were suspended separately in antibiotic saline solution in the ratio of 1 : 10. In addition, bacteriologic cultures from the internal organs were made to check their freedom from bacterial contamination. 0.2 ml. from these suspensions were inoculated into the allantoic sac of each of five, 9 to 11-day-old chicken embryos.

Isolates of NDV were identified by the rapid haemagglutination (HA) test using 10% washed chicken red blood cells and the AGPT using a locally prepared ND hyperimmune serum.

Characterization of ND isolates:

The virulence of the isolates were assessed by the following methods:

- 1- Determination of pathogenicity for chicken embryos by estimation of the mean death time of the minimal lethal dose according to ANNON (1971) and HANSON (1975).
- 2- Determination of intracerebral pathogenicity index (ICPI) in baby chicks and intramuscular pathogenicity index (IMPI) in 6-week-old chickens using groups of 10 birds each. A virus dose of  $10^6$  EID<sub>50</sub> per bird was used for inoculation. Birds were observed for death, paralysis and symptoms for 12 days postinoculation. Pathogenicity indices were determined, giving death a numerical value of 3, paralysis 2, symptoms 1, and no ill effect zero (HANSON, 1959; ANNON, 1971).
- 3- Determination of intracerebral pathogenicity index (ICPI) in pigeon squabs using groups of 10 birds per isolate. A virus dose of  $10^6$  EID<sub>50</sub> per squab was used for inoculation. The index was determined by giving death a numerical value of 4, illness 2, and no ill effect zero for an observation period of 14 days (OLAH and PALATKA, 1963).
- 4- Determination of heat stability of the haemagglutinin. This was carried out according to HANSON *ET AL.* (1949). An HI titre of 1 : 8 or higher was considered positive.

Virus reisolation:

Individual cloacal swabs were taken from experimentally infected sick and dead birds, suspended 1 : 10 in antibiotic-saline solution and inoculated into the allantoic sac of five embryonated chicken eggs. Amnioallantoic fluids were subjected to HA and HI tests.

## RESULTS

The doves showed no signs of ill health throughout the observation period.

HI antibodies to NDV were detected in 37 out of 200 apparently healthy doves (18.5%). The titres varied from 1/4 to 1/64 as shown in Table (1). Precipitins were detected in 16 doves (8%), and 12 cases showed both HI and precipitating antibodies.

TABLE (1)

Results of examination of doves sera for HI and precipitating antibodies to ND virus

	HI Antibody titre:							AGPT	
	0	1/2	1/4	1/8	1/16	1/32	1/64	+	-
No. of reactors	163	0	11	16	6	3	1	16	184

## DOVES AS CARRIERS TO NEWCASTLE DISEASE VIRUS

Two isolates of ND virus were recovered at the first egg passage level from the internal organs as well as the cloacal swabs from two apparently healthy doves. Both doves sera were positive for precipitating antibodies and had HI antibody titre of 1/8 and 1/16. The results of their identification are illustrated in Table (2).

Results of intracerebral inoculation of one-day-old chicks and pigeon squabs as well as intramuscular inoculation of 6-week-old chickens are given in Table (3). Results clearly show that both isolates are rapidly lethal to chick embryos and are of high pathogenicity for day-old chicks and 6-week-old chickens and pigeon squabs.

TABLE (2)

Results of identification of two isolates of ND virus.

Isolate	Passage	MDT/ MLD (hours)	ELD <sub>50</sub> / 0.1ml. (10g 10)	HAT	HIT	AGPT	Heat stab of haemagg. at 56 C (minutes)
I	1st	=	=	1/40	+	+	=
	2nd	=	=	1/160	+	+	=
	3rd	64	10 <sup>7.5</sup>	1/320	+	+	stab. for 30
II	1st	=	=	1/80	+	+	=
	2nd	=	=	1/160	+	+	=
	3rd	56	10 <sup>7</sup>	1/160	+	+	stab. for 30

MDT/MLD = Mean death time of minimal lethal dose.

HAT = Haemagglutination test.

HIT = Haemagglutination-inhibition test.

AGPT = Agar-gel precipitin test.

= = Not done.

TABLE (3)

Results of experimental infection with two ND virus isolates from doves.

Birds infected	Route	No. of birds	Pathog. index		Virus reisolation & HA titre			
			Isolate		Isolate			
			I	II	I	II	I	II
Day-old chicks	1/C	10	1.7	1.85	+	1/14	+	1/40
6-week-old chicken	1/m	10	2.1	2.33	+	1/160	+	1/80
Pigeon squabs	1/C	10	3.1	3.00	+	1/40	+	1/80

## DISCUSSION

The available literature show that doves are more resistant to ND virus infection than fowls (KRANEVELD and MANSJOER, 1950; REUSS, 1961; MAGID ET AL., 1965). In the present study the results of screening doves sera for HI and precipitating antibodies to ND virus, together with the isolation of two virulent strains are good evidence for exposure of these birds to ND virus. Most probably these birds had undergone a symptomless infection with some birds developed detectable antibodies. No reports are available in the literature on natural outbreaks of ND among doves. MAGID ET AL. (1965) reported that doves and sparrows placed in contact with ND-infected chickens failed to contract the disease.

MAGID ET AL. (1965) could detect HI antibodies after experimental infection of doves, but failed to isolate the virus from the feces. On the other hand, KRANEVEID and MANSJOER (1950) and RUSS (1961) proved the dissemination of the virus in the droppings of experimentally infected doves.

In the present work detection of more reactors by the HI test than by the AGP test points out the higher sensitivity of HI test and/or the earlier disappearance of precipitins.

To evaluate the pathogenicity of the recovered ND virus isolates, the criteria of HANSON (1975) were adopted. From these criteria we used the MDT of MLD, ICPI for one-day-old chicks and the IMPI for 6-week-old chickens. As recommended by OLAH and PALATKA (1963), the ICPI for pigeon squabs was also used. The results obtained from these criteria may allow the categorization of the two isolates within the range of velogenic strains (OLAH and PALATKA, 1963; AHMED ET AL., 1966; AHMED and REDA, 1967; ANNON, 1971).

The isolation of virulent strains of ND virus from apparently healthy doves no doubt suggests their role in the epizootiology of ND as they act as obligatory visitors to poultry reared extensively and to unhygienically stored rations.

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