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VACCINATION AGAINST PARAMYOVIRUS TYPE 1 (With two Tables and One Figure)

By

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(Received at 7/12/1991)

التحصين ضد مرض فيروس الباراميكرو
(نوع (1) في الحمام)

كمال الزناتي ، طلبة عبد المطلب ، بخيت سالم ، مصطفى البكري

تم تحصين مجموعة من الحمام مرتين وكان الوقت بين التحصين الأول والثاني أربعة أسابيع بواسطة اللقاح الميت المحضر عن عترة فيروس الباراميكرو المعزول من الحمام مع / أو استخدام لقاحات فيروس مرض النيوكاسل الموجودة في السوق بطرق مختلفة وتم تقييم اللقاحات علي أساس اختبائي تلازن الدم المضاد واختبار تحدي المناعة بواسطة الحقن الوريدي باستخدام العترة المعزولة من الوباء . وقد أجري اختبار تلازن الدم المضاد أسبوعيا بعد التحصين الثاني واختبار تحدي المناعة بعد أربعة أسابيع من التحصين الثاني . وكانت النتائج في المجموعة الأولى المحصنة مرتين باستخدام اللقاح المحضر (عترة الباراميكرو نوع 1) ذات أجسام مناعية عالية عند الأسبوع الثالث من التحصين الثاني وكذلك أعطت حماية أكثر في اختبار تحدي المناعة بالمقارنة بالمجموعات المحصنة الأخرى .

SUMMARY

Pigeons were vaccinated twice four weeks apart with prepared oil emulsion (OE) pigeon isolate paramyxovirus type 1 (PMV-1) and/or standard Newcastle disease virus vaccines by different systems. Evaluation of immune response was based on estimation of the hemagglutination inhibition (HI) antibody response and protection against intravenous (IV) challenge with PMVI (a field pigeon isolate). The HI titres were measured weekly post second vaccination, while challenge was done four weeks after the second dose of vaccination. Two doses of prepared OE-PMVI vaccine (pigeons in 1st group) gave higher antibody response (mean log² 9.9) at third week after the second vaccination and more protection from IV challenge in comparison with other vaccinated groups.

INTRODUCTION

Paramyxovirus type 1 infection of pigeons causing a disease resembling the neurotropic form of Newcastle disease (ND) in chickens spreaded across Europe during 1983 (ALEXANDER *et al.*, 1984 and JORDAN, 1990). The disease was recorded in many

parts of the middle east, in Isreal; WEISMAN *et al.*, 1984; SUDAN; EISA and OMER, 1984 and in upper Egypt, EL-ZANATY *et al.*, 1988.

Inactivated and live standard ND virus vaccines were effective for immunization of pigeons with different degrees of protection (VIAENE *et al.*, 1983; SOLYOM *et al.*, 1984; KALETA *et al.*, 1985 and ALEXANDER *et al.*, 1986).

The present study was carried out to determine the efficacy of locally prepared PMB 1 pigeon isolate vaccine in comparison with standard ND vaccines of chickens by different systems of vaccination.

MATERIAL and METHODS

Chicken embryos :

9 to 11-day-old embryonated chicken eggs (ECE) were supplied by the farm of faculty of agriculture, Assiut University and used in virus propagation and titration.

Pigeons :

Pigeons 4-5 week-old were obtained from the local market reared in strict isolation and used in experiments. All pigeons were negative for PMV 1-HI antibodies before the experiments.

Virus :

PMV-1 pigeon isolated previously isolated and characterized (EL-ZANATY *et al.*, 1988) was firstly inoculated in 4 pigeons under experimental control. The clinical signs, and mortality have been the same as in natural infection and the virus was isolated from diseased and dead birds, titrated in ECE ($10^{7.1}$ EID₅₀ /0.1 ml) and used for antigen preparation and challenge test.

Vaccines :

Hitchner BI : NDV live vaccine was obtained from a commercial source (TAD, CUXHAVEN) in 1000 doses vials ($10^{8.2}$ EID₅₀) titrated in ECE for efficacy before use. Pigeons vaccinated in an amount of virus equivalent to a single dose recommended for chickens (0.1 ml/pigeon, intraocularly).

Oil emulsion inactivated vaccine: It was obtained from a commercial source (Nuova Eurobio Vaccini, S.R.L. Macclodio-BS., Italy). The vaccine injected I/M 0.5 ml/pigeons as recommended for chickens.

Reference serum: A local prepared PMV-1 hyperimmune serum was used.

Antigen and vaccine preparation:

PMV-1 pigeon isolate was cultivated in ECE allantoic fluid was pooled (HA titre 640, EID₅₀ $10^{7.1}$ /0.1 ml) and inactivated with 0.1% betaprobolactone at room temperature for 4 hours.

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Inactivation of virus was confirmed by chicken embryo inoculation. Vaccine preparation was done as described by STONE *et al.*, 1978.

Haemagglutination (HA) test:

The slide and tube HA test was done after ANON, 1971.

HI test:

The microtechnique was carried out after ALLAN and GOUCH, 1974 using doubling dilutions, 0.75 percent chicken red blood cells, 4 HA units of inactivated PMV-1 pigeon isolate and 0.025 ml volumes. Titres were expressed as \log_2 of the highest dilution of serum causing complete inhibition of HA character.

Vaccination:

Sixty pigeons 6-weeks old were divided into five equal isolated groups (A-E) each containing 12 birds. Pigeons of each group were vaccinated twice as shown in table 1. The apart between the first and second vaccination was four weeks. Sera were collected weekly for four weeks after the second vaccination for determining HI-antibodies.

Challenge test:

Pigeons of each group were challenged I/V four weeks after the second vaccination with locally isolated PMV-1 of pigeons (EID₅₀ $10^{7.1}$ / 0.1 ml/pigeon). This route was previously known to produce nervous signs and deaths in susceptible pigeons similar to those seen in the field outbreak. All pigeons were observed for four weeks post challenge and any clinical signs or deaths were recorded. Trials for virus reisolation from diseased (cloacal swabs) or from internal organs (brain, liver, spleen) of dead birds were adopted.

RESULTS

The serological responses induced by different systems of vaccination in pigeons are shown in Fig. 1. The highest HI-antibodies was recorded in the vaccinated pigeons of the first group at three weeks after the second vaccination (\log_2 8.9), while at the second week after the second vaccination, the HI-antibodies mean \log_2 was higher in the third group (\log_2 7.1) followed by the first group (\log_2 6.8) as shown in Fig.1.

Challenge tests revealed that the most protection was observed in the pigeons of the first group and the lowest protection in pigeons of the fourth group (Table 2). The clinical signs were depression, inappetence, diarrhoea, incoordination, partial to complete paralysis and torticollis. The onset of the clinical signs were illustrated in Table 2. The earliest clinical signs were observed in the third and fourth vaccinated challenged pigeons groups. Some of diseased pigeons showing only depression, diarrhoea were recovered, while pigeons with nervous signs were usually die.

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No deaths occurred in the pigeons of the first group but in the other groups, deaths recorded between 6-15 days post-challenge Table 2.

Virus reisolation was successful from cloacal swabs of sick pigeons and from internal organs of dead ones.

Table (1) : Vaccination of pigeons with prepared OE PMV1 (pigeon isolate and standard ND vaccines.

Vacc. group	First vaccination		Second vaccination	
	Type of vaccine	Route	Type of vaccine	Route
1st	prep. OE	S/C	prep. OE	S/C
2nd	Comm. OE	S/C	Comm. OE	S/C
3rd	HB1	I/O	prep. OE	S/C
4th	HB1	I/O	Comm. OE	S/C
5th	Control non. vaccinated			

prep. OE = prepared oil emulsion vaccine.
 Comm. OE = commercial oil emulsion vaccine.
 HB1 = Hitchner B1 vaccine.
 S/C = subcutaneous
 I/O = intra-ocular

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Table 2: Challenge of vaccinated pigeons with PMV 1 pigeon isolate in different groups in addition to control nonvaccinated group.

Vacc. group	Sick pigeons															Dead pigeons																		
	Onset of clinical signs in days post-challenge															Daily deaths post-challenge																		
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	No.	%	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	No.	%
1st	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1/12	8.3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0/12	0.0
2nd	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	3/12	25.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1/12	8.3
3rd	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4/12	33.3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2/12	16.7
4th	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4/12	33.3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	3/12	25.0
5th	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	12/12	100.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	7/12	58.3

+ = One pigeon

2+ = Two pigeons

Fig. 1: HI mean log₂ titre of vaccinated pigeons in different groups weekly after the second vaccination

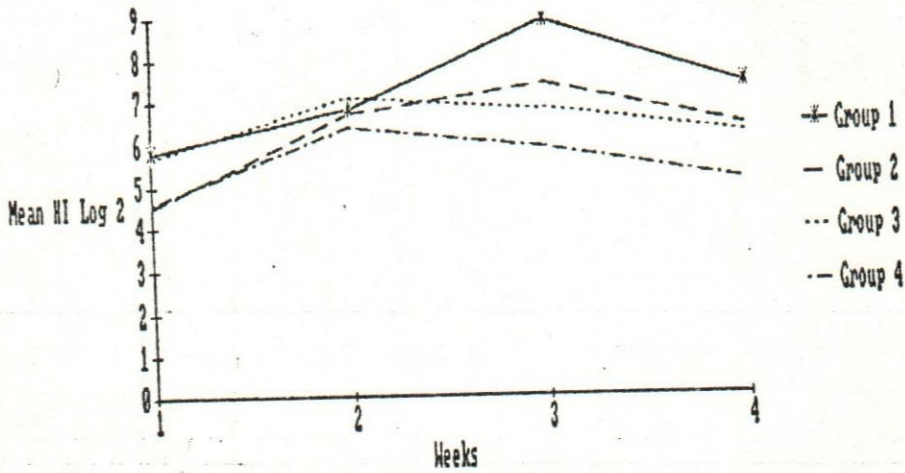


Fig. 1: HI mean log₂ titre of vaccinated pigeons in different groups weekly after the second vaccination



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DISCUSSION

In the present study, the serological experiments indicated that vaccination of pigeons with two doses of either prepared inactivated PMV-1 or commercial ND vaccine gave better HI-antibodies mean titre and more protection than those initially vaccinated with living ND-HB₁ vaccine and followed by inactivated OE (prepared PMV-1 or commercial ND) vaccine. KALETA *et al.*, 1985 reported that the inactivated vaccines were superior to the live vaccines in their ability to induce demonstrable HI antibody titres. Our results are agreed with ALEXANDER *et al.*, 1986 in that more protection was obtained with two doses of OE inactivated vaccine than those vaccinated with live ND-HB₁ and followed by inactivated vaccine. Vaccination of pigeons with two doses of prepared inactivated OE PMV-1 vaccine produce more protection than the commercial ND vaccine, similar results were reported by KNOLL *et al.*, 1986. This may be due to the antigenic variation between PMV-1 pigeons isolate and the classical ND virus strains. The peak HI-antibodies mean titre in vaccinated groups (first and second) reached at three weeks after the second vaccination which disagreed with BOX *et al.*, 1985 who reported that good antibody response was two weeks after the second vaccination.

Endly, it is clear that better protection was obtained in pigeons vaccinated with two doses of prepared inactivated OE PMV-1 vaccine than those vaccinated with either living and inactivated OE (prepared or commercial) or two doses of inactivated OE classical ND vaccines. Frequent Vaccination is also recommended because HI titres decreased three weeks after the second vaccination

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