

STUDIES ON VACCINATION OF EGYPTIAN ZOO DUCKS BY LOCAL INACTIVATED DUCK CHOLERA AND DUCK HEPATITIS VACCINES IN EGYPT

By

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SUMMARY

*As an interesting subject; some trials were carried out to investigate the immune response of zoo ducks to duck hepatitis (DH) and duck cholera (DC) inactivated local vaccines. It was found that vaccinated dams responded well to the simultaneous vaccination with duck hepatitis and duck cholera inactivated vaccines showing high titers of specific antibodies against both of DH and DC without any antagonizing effect between them. These dams allowed their offspring to be immune against the two diseases through the exhibited maternal immunity where such ducklings were able to withstand the challenge with virulent strains of DVH and DC up to 2 weeks of age. On the other hand vaccinated ducklings at 2 days of age with DH vaccine exhibited high levels of neutralizing antibodies that enable them to overcome the challenge with the virulent virus. Also DC vaccine was able to induce high levels of DC antibodies in vaccinated birds and protected them against the challenge with virulent *P.multocida* strain. In addition, it was found that both of DH and DC vaccines were safe either in single or simultaneous administration for ducks and ducklings.*

INTRODUCTION

Wild birds are used as in captive birds in zoo gardens of which there are several ones in Egypt. Birds in Egypt either they are domestic or wild belong to 71 families and 12 orders representing 515 species and sub-species (Tharwat, 1997).

Both wild and domestic ducks are susceptible to many bacterial and viral infectious diseases which affect them dramatically causing great economic losses. Among these diseases that threaten duck breeding are duck cholera and duck hepatitis which constitute a major cause of high mortality in duck farms (Heddleston, 1972 and Levine, 1972).

Duck cholera (DC) is a highly contagious bacterial disease affecting wild and domestic ducks and geese. DC is considered one of the most important diseases affecting duck populations (Ghazikhanian et.al., 1983 and

Sander et.al., 1998). DC in water fowls is usually an acute or peracute disease characterized by septicaemia and focal necrosis with bacterial colonies in the liver, spleen and other organs (Hunter and Wobeser, 1980). The causative agent of DC is *Pasteurella multocida* serotype A (Jonas et.al., 2001 and Rocke et.al., 2002).

These diseases may appear to be the most fatal diseases facing ducklings and they have the ability to destruct completely a duck colony. Such dramatic effect can cause great economic losses especially among zoo ducks and accordingly the main aim of the present study is to evaluate the immune response of zoo ducks to simultaneous vaccination with the locally produced duck cholera and duck hepatitis vaccines.

MATERIAL AND METHODS

1-Vaccines:

1.1-Duck hepatitis vaccine:

Inactivated duck hepatitis vaccine was supplied by Veterinary Serum and Vaccine Research Institute, Abassia, Cairo. It was used for vaccination of experimental ducklings at a dose of 0.1ml/bird (of 10^7 EID₅₀ DH virus/ml) inoculated S/C.

1.2-Duck cholera vaccine:

Inactivated duck cholera vaccine was supplied by Veterinary Serum and Vaccine Research Institute, Abassia, Cairo. It was used for vaccination of experimental ducklings at the dose of 0.5ml/bird inoculated S/C and a 2nd dose was administered 2 weeks later.

2-Virus strains:

2.1-Virulent duck hepatitis virus:

A local strain of DH virus (Mervat, 1997) of a titer $10^{8.58}$ EID₅₀/ml was kindly supplied by the Central laboratory for Evaluation of Veterinary biologics; Abassia; Cairo. It was used for challenge of vaccinated ducklings at a dose of 0.1ml containing 10^7 EID₅₀ /duckling inoculated I / M according to Mervat et.al. (1999).

2.2-Cell culture adapted DH virus:

DH virus at its 11th passage on VERO cells of a titer 10^8 TCID₅₀/ml (Mervat, 1997) was used for estimation of DH antibodies using serum neutralization test.

3-Virulent duck cholera strain:

A local strain of virulent *Pasteurella multocida* serotype A:1 was kindly supplied by the Central laboratory for Evaluation of Veterinary biologics; Abassia; Cairo. It was used for challenge of vaccinated ducklings.

4-Serum neutralization test (SNT):

SNT was carried out to estimate DH neutralizing antibodies in vaccinated birds using the micro titer technique according to Kaleta (1988). The neutralizing antibody titer was calculated as the reciprocal of the final

serum dilution which neutralized and inhibited the cytopathic effect (CPE) of 100-200 TCID₅₀ of DH virus according to **Singh et.al. (1967)**.

5-Indirect haemagglutination test (IHA):

It was carried out to estimate DC antibodies in vaccinated ducks according to **Carter and Rappy (1962)**.

RESULTS AND DISCUSSION

It is well known that vaccination against infectious; diseases either bacterial or viral diseases; is the most effective measure and is the corner stone in controlling such diseases. Zoo ducks as other zoo birds could be considered of a public wealth importance and accordingly they must be protected against infectious diseases. So the present work was directed to study the immune response of zoo ducks and ducklings to the locally produced inactivated duck hepatitis and duck cholera vaccines.

The experimental results showed that vaccinated dams simultaneously with the two vaccines, did not show any signs of illness and exhibited detectable antibodies against both of DH and DC without any antagonizing effect between them by the 1st week post the 1st dose then increased after the 2nd dose to record their highest titers by the 3rd week after the 2nd vaccination (5th week post the 1st vaccination). These dams were able to withstand the challenge with virulent DH and DC strains and transmitted such immunity to their offspring protecting them against the challenge with virulent strains. Concerning DH; serum neutralization test (Tab-1) revealed that the detected levels of antibodies could be considered of high protective values and these results are in agreement with those obtained by Nikitin and Panikar (1974); Fan et.al. (1993); Mervat (1997) and Mervat et.al. (1999)

On the other hand vaccination of ducklings from unvaccinated dams; with inactivated DH vaccine exhibited good protective levels of neutralizing antibodies by the 1st week post vaccination and increased to reach a peak titer by the 3rd week post the 2nd dose enabling birds to withstand the challenge with virulent DH virus. These results are in agreement with the findings of Nikitin and Panikar (1974) and Bezrukavaya (1978) who confirmed that 2 or 3 doses of inactivated DH vaccine were necessary to secure satisfactory levels of protection. Also Davis and Hannant (1986) detected DH neutralizing antibodies in the sera of vaccinated ducklings 4 days post vaccination. It was found that all vaccinated ducklings and ducklings with maternal immunity of a titer 64 were able to overcome the infection with the virulent DH virus (with 100% protection rate) while the unvaccinated ducklings and those with lower maternal antibodies were not (showing 0% protection rate and characteristic signs of duck hepatitis) in agreement with Golubnich and Malinovskaya (1984) Mervat (1997) and Mervat et.al. (1999).

On the other side, it was found that the inactivated DC vaccine was able to induce detectable antibodies in vaccinated birds as demonstrated by the indirect haemagglutination test (Table-2) by the 1st week post the dose of vaccination and increased by the 2nd dose recorded their highest level 3 weeks post the 2nd vaccination. Also the offspring of vaccinated ducks exhibited maternal immunity of good protective levels up to 2 weeks of age. It was found that all vaccinated birds and ducklings with good maternal immunity were able to withstand the challenge with the virulent *P.multocida* type A:1 while the unvaccinated ducks and ducklings with low or without maternal immunity was unable to withstand such challenge (showing typical DC signs and 90% deaths). Similar results were obtained by Rice et.al. (1978); Schlink and Olson (1987); Swamy et.al. (1996); Kedrak et.al. (2000) and Hanan (2004) who demonstrated that *P.multocida* inactivated vaccines induce antibodies of high HI titer in the sera of vaccinated ducks and ducklings.

From the obtained results it could be concluded that the locally produced inactivated DH and DC vaccines are suitable for vaccination of zoo duck breeders and ducklings providing them with good immunity saving them against the most serious disease facing domestic and wild duck population.

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Table (1): Mean titer of DH neutralizing antibodies in zoo ducks and ducklings

| Bird group | Mean titer* of DH neutralizing antibodies/WPV** | | | | | |
|---|---|---------|------|------|------|------|
| | Prevac.*** | 1WPV | 2WPV | 3WPV | 4WPV | 5WPV |
| Dam ducks | 0 | 4 | 8 | 32 | 64 | 128 |
| Ducklings from vaccinated dams | Maternal immunity | 1WA**** | 2WA | 3WA | 3WA | 4WA |
| | | 64 | 16 | 4 | 0 | 0 |
| Vaccinated ducklings from unvaccinated dams | 0 | 8 | 32 | 64 | 128 | 128 |
| Unvaccinated ducklings | 0 | 0 | 0 | 0 | 0 | 0 |

*Antibody titer= the reciprocal of the final serum dilution which neutralized and inhibited the CPE of 100-200 TCID₅₀ of DH virus.

WPV= Week post vaccination *Prevac. = Pre vaccination

****WA= Week of age

Table (2): Monitoring of DC antibodies in the sera of zoo ducks and ducklings

| Bird group | Indirect haemagglutination titers/weeks post vaccination | | | | | |
|---|--|--------|------|------|------|------|
| | Prevac.* | 1WPV** | 2WPV | 3WPV | 4WPV | 5WPV |
| Dam ducks | 8 | 140 | 280 | 453 | 520 | 597 |
| Ducklings from vaccinated dams | Maternal immunity | 1WA*** | 2WA | 3WA | 3WA | 4WA |
| | | 98 | 25 | 8 | 2 | 0 |
| Vaccinated ducklings from unvaccinated dams | At 4 weeks of age | 125 | 260 | 368 | 520 | 640 |
| | 8 | | | | | |
| Unvaccinated ducklings | 2 | 2 | 4 | 2 | 4 | 4 |

*Prevac. = Pre vaccination

**WPV= Week post vaccination

***WA= Week of age

الملخص العربي

دراسات على تحصين بط حديقة الحيوان المصرية بلقاح كوليرا البط والالتهاب الكبدى الميت

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*المعمل المركزى لرقابة على المستحضرات الحيوية البيطرية-العباسية

**معهد بحوث الأمصال واللقاحات البيطرية-العباسية- القاهرة

شملت الدراسة الحالية مجموعة من إناث وذكور بط حدائق الحيوان بالجيزة تم التأكد من خلوها من الأجسام المناعية لكل من كوليرا البط والالتهاب الكبدى للبط تم تحصين مجموعة من الإناث تزامنيا بلقاحى كوليرا البط والالتهاب الكبدى المثبتين والمنتجين محليا وتركت مجموعة أخرى من الإناث دون تحصين وبالحصول على بيض مخصب من كلا المجموعتين تم الحصول على مجموعتين من البط الصغير أحدهما بها مناعة مكتسبة من الأمهات والأخرى خالية من هذه المناعة وهى التى تم تحصينها بلقاح الالتهاب الكبدى عند عمر يومين وبلقاح كوليرا البط عند عمر أربعة أسابيع حيث اعطيت جرعتين من كل لقاح بفترة بينية أسبوعين 0 وقد أوضحت نتائج أختبارات المصل المتعادل ومانع تلازن الدم الغير مباشر أن الطيور المحصنة سواء الأمهات أو الصغار تكتسب مستويات مناعية عالية ضد كل من الكوليرا والالتهاب الكبدى تصل ذروتها فى الأسبوع الثالث بعد الجرعة الثانية وتمكن هذه المناعة الطيور المحصنة من التغلب على العدوى التجريبية بالعترات الضارية 0 أما بالنسبة لصغار البط الناتج من أمهات محصنة فقد وجد أنها تكتسب مناعة ذات مستوى جيد تمكنها من التغلب على التحدى بالعترت الضارية للكوليرا والالتهاب الكبدى وحتى أسبوعين من العمر الأمر الذى يبين مدى أهمية تحصين هذه النوعية من البط ولاسيما الأمهات بلقاحى الكوليرا والالتهاب الكبدى حتى يمكن حماية هذه الطيور من هذين المرضين اللذين يعتبران من أخطر الأمراض التى تصيب البط سواء المستأنس أو البرى حفاظا على تلك الثروة القومية