Bangl. J. Vet. Med. 2008. 6 (1): 27–30

STANDARDIZATION OF AGE AND ROUTE FOR DUCK PLAGUE VACCINE IN LOCAL DUCKLINGS OF VACCINATED AND NON- VACCINATED PARENT ORIGIN

M. E. H. Kayesh[1], M. S. R. Khan[1], M. A. Islam[1], M. O. Gani[3], M. R. Islam[1], M. R. Karim[1], M. S. Islam[2] and A. Kabir[2]

[1]Department of Microbiology and Hygiene, [2]Department of Medicine, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh, [3]Goat and Sheep Production Research Division, BLRI, Savar, Dhaka, Bangladesh

ABSTRACT

The study was carried out to determine the appropriate age and route of vaccination with duck plague vaccine (LRI, Mohakhali) in experimentally reared local ducks during the period from September 2006 to May 2007. A total of 90 local ducklings were divided into eight groups namely A, C, D and E for vaccinated parent origin and B, F, G and H for non-vaccinated parent origin. Again Group C, D, E, F, G and H were divided into subgroups C_1 & C_2; D_1& D_2; E_1& E_2; F_1 & F_2; G_1 & G_2; H_1 & H_2 respectively and contained 5 ducklings each. Group A and B contained 15 ducklings in each group and were used as unvaccinated control and also for studying persistence of maternally derived antibody level. Highest mean MDA titre in ducklings of Group A was found 53.33 ± 4.03 at the age of day 1 that declined to a negligible level (≤4) at the age of day 21 and highest mean MDA titre in ducklings of Group B was found 29.86 ± 1.45 at the age of day 1 that declined to a negligible level (≤4) at the age of day 16. Ducklings of different sub-groups were vaccinated with duck plague vaccine at day 14, day 21 and day 28 through intramuscular (breast) and subcutaneous route at the dose rate of 1ml per duckling. Sera were collected from vaccinated ducklings on day 7, day 14 and day 21 after vaccination and antibody titre was measured by passive hemagglutination test. Among the vaccinated subgroups, the highest mean PHA titre was found 89.60 ± 15.67 and 83.20 ± 19.20 in subgroups E_1 (vaccinated at day 28) and F_1 (vaccinated at day 14) respectively. At challenge, ducklings of vaccinated subgroups showed 100% protection except C_1 and C_2 (showed 80% protection) and control subgroups showed 0% protection. From the results of protection test it may be concluded that both intramuscular (breast) and subcutaneous routes are equally suitable for duck plague virus vaccination in ducklings and the optimum age for vaccination to ducklings originated from vaccinated parent to duck plague vaccine might be at day 21 or 28 and ducklings originated from non-vaccinated parent at day 14 or 21 instead of usual schedule of day 28.

Key words: Vaccination, duck plague vaccine, age, route, ducklings

INTRODUCTION

Though ducks are considered relatively resistant birds compared to other members of domestic poultry, infection caused by duck plague virus is important for all age groups of ducks which is characterized by high morbidity and mortality varying from 5-100% (Calnek *et al*., 1997). The disease frequently occurs every year in Bangladesh in epidemic form and spreads rapidly among the duck population of the duck raising areas. The causal agent was first isolated in Bangladesh by Sarker, 1980. About 60-70% duck mortality occurs due to duck plague in Bangladesh (Sarker, 1982). In Bangladesh, DPV vaccine produced at Livestock Research Institute (LRI), Mohakhali is reported to provide a good immunity in protection of DP infection. But it is also reported that sometimes this vaccine fails to protect the ducks despite regular vaccination (personnel communication). The reason might be due to improper timing of vaccination and persistence of MDA in individual ducklings. Proper vaccination is the most effective means of controlling duck plague virus infection. Leonchuk and Tsimopkh (1977) reported that the immunogenicity of vaccine depends on the method of vaccination and the amount of doses given. For effective control of DP, proper age of duck due to the presence or absence of maternally derived antibody, the route of vaccination and dose of vaccine must be considered before vaccination. Therefore, the present study was undertaken to determine the appropriate age and route of vaccination of duck plague vaccine for the refinement of vaccination schedule and also to study the efficacy of duck plague vaccine in relation to age and route in experimentally reared local ducks.

MATERIALS AND METHODS

The study was conducted in the Department of Microbiology and Hygiene, Faculty of Veterinary Science, BAU, Mymensingh during the period from September 2006 to May 2007. The local virulent DPV isolate was obtained from the laboratory repository of the Department of Microbiology and Hygiene, BAU, Mymensingh. For reactivation, the virus was given 8 passages in embryonated duck eggs. The virus was propagated through chorio-allantoic membrane (CAM) route in 10 days old duck embryos and this virus isolate was used for passive hemagglutination (PHA) test and challenge test.

Duck eggs were purchased locally and from BAU Poultry Farm, Mymensingh and were used for hatching ducklings and also for the cultivation of DPV. A total of 90 local ducklings were used for this study. Lyophilized duck plague vaccine produced at Livestock Research Institute (LRI), Mohakhali, Dhaka, Bangladesh was used to vaccinate the ducklings. All the ducklings were reared in a well-ventilated duck house of the Department of Microbiology and Hygiene, BAU, Mymensingh, providing feed and water *ad libitum*.

For MDA titre determinations, ducklings of both vaccinated and non-vaccinated parent origin were divided into two groups namely group A and B contained 15 ducklings each. Sera were collected from ducklings at day 1, 6, 11, 16 and 21 of age and antibody titre was determined by PHA test as per method described by Zyambo *et al.* (1973) with some modifications described by Tripathy *et al.* (1970). Remaining ducklings were divided into six groups and were again subdivided into two sub-groups each namely C_1 & C_2, D_1 & D_2, E_1 & E_2, F_1 & F_2, G_1 & G_2, H_1& H_2 where C_1, D_1, E_1, F_1, G_1 and H_1 were selected to be vaccinated through intramuscular breast route (IMB) and C_2, D_2, E_2, F_2, G_2 and H_2 were selected to be vaccinated through subcutaneous route (SC) at the dose rate of 1 ml per duckling. Ducklings of sub-groups C_1, C_2, F_1 & F_2; D1, D_2, G_1 & G_2 and E_1, E_2, H_1 and H_2 were vaccinated on day 14, 21 and 28, respectively. Blood was collected for sera from each sub-group on day 7, 14 and 21 of post vaccinations to determine the immune response against DPV vaccine. Antibody titres of sera samples were determined by PHA test.

For protective efficacy study $DELD_{50}$ was determined and the calculation was done according to the methods described by Reed and Muench (1938). All the vaccinated and non-vaccinated parent origin ducklings were challenged with 1 ml of 100 $DELD_{50}$ virulent DPV isolate through IM route after 21 days of vaccination. After challenge exposure all the ducklings were kept in separate room and observed up to 10 days for the development of clinical signs and symptoms and for death of the individual.

RESULTS AND DISCUSSION

The results of MDA titres in ducklings originated from vaccinated and non-vaccinated parent to DPV vaccine from day 1 to day 21 are presented in Table 1. It was observed that, highest mean MDA titre in ducklings of Group A was found 53.33 ± 4.03 at the age of day 1 that declined to a negligible level (≤4) at the age of day 16 and highest mean MDA titre in ducklings of Group B was found 29.86 ± 1.45 at the age of day 1 that declined to a negligible level (≤4) at the age of day 11.

Table 1. Maternally derived antibody titre of ducklings originated from vaccinated and non-vaccinated parent

Age (day)	MDA titre in ducklings of vaccinated parent origin (mean ± SE of mean)	MDA titre in ducklings of non-vaccinated parent origin (mean ± SE of mean)
Day 1	53.33 ± 4.03	29.86 ± 1.45
Day 6	36.26 ± 2.90	18.13 ± 1.45
Day 11	23.46 ± 2.13	≤4 ± 0
Day 16	≤4 ± 0	≤4 ± 0
Day 21	≤4 ± 0	≤4 ± 0

≤ = Less than or equal to; DPV vaccine = Duck plague virus vaccine.

Toth and Suwathanaviroj (1979) mentioned that ducklings less than two weeks of age did not require vaccine because of maternally derived antibody that protects them from DPV infection. Kumar and Punnose (2000) stated that PHA titre 22 ± 0.7 containing ducklings showed 100% protection on challenge with virulent DPV. It was observed that, MDA titre persisted in protective level up to day 11in ducklings of group A, whereas in ducklings of group B titre goes below protective level at day 6. From the above findings, it can be said that ducklings from vaccinated parent origin should not be vaccinated before two weeks of age, because of presence of high level of MDA titre may interfere in the development of sufficient antibody titre after vaccination with DPV vaccine.

In Bangladesh, DPV vaccine usually administered through IM route but no report of using SC route. For this reason, the study was undertaken to know the efficacy of DPV vaccine through SC route and to justify the feasibility of using DPV vaccine through SC route as an alternate of usual IM route.

In case of intramuscular vaccination, among the sub-groups originated from vaccinated parent the highest mean antibody titre was found 89.60 ± 15.67 in sub-group E_1 (vaccinated at day 28) and among the sub-groups originated from non-vaccinated parent the highest mean antibody titre was found 83.20 ± 19.20 in sub-group F_1(vaccinated at day 14) (Table 2). In case of subcutaneous vaccination, among the sub-groups of vaccinated parent origin the highest mean antibody titre was found 83.20 ± 19.20 in sub-group E_2 (vaccinated at day 28) and among the sub-groups of non-vaccinated parent origin the highest mean antibody titre was found 76.80 ± 12.80 in sub-group F_2 (vaccinated at day 14) (Table 2).

Table 2. Mean PHA titre in ducklings originated from vaccinated and non-vaccinated parent to DPV vaccine

Sub-groups	Route of vaccination	Age of vaccination (day)	Pre-vaccination titre	Mean PHA titre (Mean± SE) after vaccination		
				7 days	14 days	21 days
C_1	IBM	14	12.80± 1.95	19.20 ± 3.20	25.60 ± 3.91	38.40 ± 6.40
C_2	SC		12.80± 1.95	16.00 ± 0.00	22.40 ± 3.91	35.20 ± 7.83
D_1	IMB	21	≤ 4 ± 0	22.40 ± 3.91	51.20 ± 7.83	64.00 ± 17.52
D_2	SC		≤ 4 ± 0	19.20 ± 3.20	44.80 ± 7.83	57.60 ± 6.40
E_1	IMB	28	≤ 4 ± 0	25.60 ± 3.91	57.60 ± 6.40	89.60 ± 15.67
E_2	SC		≤ 4 ± 0	22.40 ± 3.91	51.20 ± 7.83	83.20 ± 19.20
F_1	IMB	14	6.40± 0.97	22.40 ± 3.91	57.60 ± 6.40	83.20 ± 19.20
F_2	SC		6.40± 0.97	19.20 ± 3.20	51.20 ± 7.83	76.80 ± 12.80
G_1	IMB	21	≤ 4 ± 0	28.80 ± 3.20	51.20 ± 7.83	76.80 ± 12.80
G_2	SC		≤ 4 ± 0	25.60 ± 3.91	48.00 ± 10.11	70.40 ± 15.67
H_1	IMB	28	≤ 4 ± 0	22.40 ± 3.91	44.80 ± 7.83	64.00 ± 17.52
H_2	SC		≤ 4 ± 0	19.20 ± 3.20	41.60 ± 9.60	57.60 ± 6.40

IMB = Intramuscular breast, SC = Subcutaneous, SE = Standard error, ≤ = Less than or equal to, PHA = Passive haemaglutination test.

The results of survivability rate are presented in Table 3. It was found that one duckling from each sub-group C_1 and C_2 died within 10 days of challenge exposure. It might be due to individual variations in duck. All the ducklings of control groups A and B died within 10 days of challenge exposure. The survivability rates in case of sub-groups D1, D_2, E_1, E_2, F_1, F_2, G_1, G_2, H_1 and H_2 were 100%, whereas survivability rate in sub-groups C_1 and C_2 was 80%. It was observed that the development of antibody titre after vaccination with DPV vaccine was satisfactory in both the routes. As it was found that ducklings of sub-group C_1 and C_2 did not show 100% protection, this might be due to the presence of high MDA titre in that individual ducklings at the time of vaccination with DPV vaccine which interfered to develop the antibody titre at high protective level.

Table 3. Survivability rate after challenge in ducks of all groups after 21 days of post vaccination

Sub-groups	Age of vaccination (day)	Age of challenge (day)	No. of ducklings	No. of ducklings dead	No. of ducklings survived	Survivability rate (%)
C_1	14	35	5	1	4	80
C_2	14	35	5	1	4	80
D_1	21	42	5	0	5	100
D_2	21	42	5	0	5	100
E_1	28	49	5	0	5	100
E_2	28	49	5	0	5	100
A	Not done	35	5	5	0	00
		42	5	5	0	00
		49	5	5	0	00
F_1	14	35	5	0	5	100
F_2	14	35	5	0	5	100
G_1	21	42	5	0	5	100
G_2	21	42	5	0	5	100
H_1	28	49	5	0	5	100
H_2	28	49	5	0	5	100
B	Not done	35	5	5	0	00
		42	5	5	0	00
		49	5	5	0	00

From the above findings it may be concluded that both intramuscular (breast) and subcutaneous routes are equally suitable for DPV vaccination in ducklings and the optimum age for vaccination to ducklings originated from vaccinated parent might be at day 21 or 28 and ducklings originated from non-vaccinated parent at day 14 or 21 instead of usual schedule of day 28.

ACKNOWLEDGEMENTS

The authors are extremely grateful to Muhammad Tofazzal Hossain, Assistant Professor, Department of Microbiology and Hygiene, Bangladesh Agricultural University, Mymensingh for his scholastic suggestions to complete the work.

REFERENCES

1. Calnek BW, Barnes HJ, Beard CW, McDougald LR and Saif YM (1997). *Diseases of Poultry*. 10th edn., Iowa State University Press, Ames, Iowa, USA. pp. 675-683.
2. Kumar SK and Punnose TK (2000). Immunogenicity of chicken embryo fibroblast cell culture adapted vaccine strain of duck plague virus. *Indian Veterinary Journal* 77: 89-91.
3. Leonchuk SI and Tsimopkh PP (1977). Immunological changes in chickens vaccinated with emulsified *Pasturella multocida* vaccine. *Poultry Abstract* 4: 271.
4. Reed LJ and Muench H (1938). A simple method of estimating fifty percent end points. *American Journal of Hygiene.* 27: 493-496.
5. Sarker AJ (1980). Duck plague in Bangladesh. *Indian Veterinary Journal* 57: 1-5.
6. Sarker AJ (1982). Duck plague in Bangladesh: Isolation and identification of the etiological agent. *Indian Veterinary Journal* 59: 669-679.
7. Toth B and Suwathanaviroj V (1979). Outbreak of duck plague. *Veterinary Bulletin* 49: 417.
8. Tripathy DN, Hanson LE and Mayrs WL (1970). Passive hemagglutination with fowl pox virus. *Avian Diseases* 14: 29-38.
9. Zyambo GCN, Dennet DP and Johnson RH (1973). A passive haemagglutination test for the demonstration of antibody of infectious bovine rhinotrachitis/ infectious pustular vulvovagainitis virus. I. Standardization of test components. *Australian Veterinary Journal* 49: 409-412.