



Comparative response to maternal vaccination, *in ovo* vaccination and post hatch vaccination on the neonatal immunocompetent traits of turkey poults

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ABSTRACT

An experiment was designed involving maternal vaccination (MV or no MV), *in ovo* vaccination (IV or no IV) and conventional vaccination (CV or no CV). 7 days post ND (Newcastle disease) vaccination, 460 fertile eggs were collected and on 25th embryonic day (ED), 230 eggs were *in ovo* vaccinated with formaldehyde inactivated ND F₁ vaccine. After hatch, half of the turkey chicks from respective group were conventionally vaccinated and reared. The group having maternal vaccination and *in ovo* vaccination had higher hatchability. Haemagglutination inhibition (HI) titer (log₂) against ND vaccination was significantly higher in poults hatched from maternally vaccinated hens than the un-vaccinated ones up to 28 days of age. The poults with *in ovo* vaccination, had higher titer than un-*in ovo* vaccinated poults till 49 days of age. Combination of maternal vaccination and *in ovo* vaccination had higher titre value than that *in ovo* vaccinated group till 21 days of age. Thereafter only *in ovo* vaccinated groups had higher titre than either maternal or combination of both. However, they had significantly higher titer values than unvaccinated poults. *In ovo* vaccination also had apparently higher titer values than conventional vaccination till 28 days of age. *In ovo* vaccination and conventional vaccination did not show any beneficial effect on HI titer. Only maternal vaccination maintained higher titer value than conventional vaccination up to 21 days of age. Thereafter, conventional vaccinated group had higher titer value. Combination of maternal vaccination and conventional vaccination did not elicit better immune response than either maternal or conventional vaccination. Hence, it may be concluded that *in ovo* vaccination did not affect the hatchability and combination of maternal and *in ovo* vaccination may elicit better antibody titer against ND vaccination in the progeny.

Key words: Breeder vaccination, Immunity, *In ovo* vaccination, Turkey

Over the past few years enhancing the neonatal immunity of chicks have drawn attention of scientists throughout the world. Breeder vaccination, *in ovo* vaccination and conventional vaccination at day-old are the different types of vaccination followed to elicit the immune response of the neonates. Higher antibody in day-old broilers results in few vaccine induced reactions, less vaccine virus shed and decreased duration of vaccination-induced immunity from coarse-spray vaccination (Giambrone and Closser 1990). The antibodies pass to the egg and thereby to the chicks, which have the potency to counteract the pathogens till 3 weeks. However, breeder vaccination imparts a lot of stress to the birds. Quaglio and Lombardi (1973) reported that administration of a vaccine to a chick in the presence of

significant amounts of circulating maternally derived antibody, increased the rate of removal of antibody and rendered the chick more responsive to subsequent application of vaccine. Ask *et al.* (2004) pointed out that a high maternal immunity might inhibit vaccination effects, and thereby increase health risks, when it decreases. An intermediate maternal immunity is optimal for chick health. Low natural and acquired immunity result in self-inhibiting ontogenesis of the immune system. Hence, a high natural immunity and acquired immunity is optimal (Ask *et al.* 2004).

Thus, the conventional approach to disease prevention by post hatch vaccination at times fails to impart the desired immunity. To overcome the problems of conventional vaccination *in ovo* vaccination was developed. The vaccine viruses do not adversely affect hatchability and chicks hatching from vaccine injected eggs show evidence of protective immunity at hatch. Hence, development of an *in ovo* vaccination package against different diseases in turkeys may enhance the immunocompetence of turkey neonates. However, before attempting to develop such a package, a

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comparative study is necessary to evaluate the effect of maternal vaccination, conventional vaccination and *in ovo* vaccination on the post hatch immune competence traits. Keeping this in view, the present study was carried out to evaluate the effect of different routes of vaccination on the neonatal immune competence traits.

MATERIALS AND METHODS

Birds: Turkey breeder hens (200) and viable males (24) of 30–35 weeks of age of small white variety were procured from the institute, and distributed into 2 treatment groups having 4 replicates of 25 hens and 3 toms in each treatment. They were offered turkey breeder diet (NRC 1994). The first group was vaccinated with ND (R₂B) vaccine whereas the other group remained as the non vaccinated group. The fertile eggs were collected from these hens for *in ovo* vaccination.

Collection of fertile eggs: Standard size fertilized turkey eggs (460) from each main group were collected 7 days after vaccination and divided into 2 primary sub groups. First group consisted of eggs from vaccinated breeders whereas the second group consisted of eggs from the non vaccinated breeders. On 25th embryonic day (ED), half of the eggs from each group were *in ovo* vaccinated with ND F1 strain formaldehyde killed vaccine of concentration 10³ titer at the broad end of the egg for deposition of the vaccine in the amniotic sac resulting into 4 groups: eggs from breeders vaccinated and *in ovo* vaccinated, eggs from breeders only vaccinated, eggs from non vaccinated breeders but *in ovo* vaccinated, eggs from non vaccinated breeders. The turkey poults hatched from each of the 4 groups (2 breeder vaccinations × 2 *in ovo* vaccinations) were further divided into 2 secondary sub groups during post hatch period: conventionally vaccinated with ND F1 strain vaccine and non-vaccinated and reared up to 8 weeks of age to study growth performance and immune status of the poults. The chicks were fed ration having 28% CP and 3000 ME/kg up to 8 weeks of age.

***In ovo* vaccination:** Based on results obtained from preliminary standardization experiments conducted in our laboratory, the eggs were injected by a 25 mm 24 gauge needle at the broad end of the egg at 25th ED with Newcastle disease vaccine (F1) strain of concentration 10³ titer killed with 1% formaldehyde.

Immune response parameters studied: Specific immune response to ND vaccine (both F1 and R2B) was studied by Hemagglutination inhibition (HI) method as described by Allan and Gough (1974) at 0, 7, 14, 21, 28, 42 and 49 days (d), respectively, by collecting blood from 6 birds from each treatment group.

Statistical analysis: All the data obtained were analyzed as per the standard statistical procedure (Snedecor and Cochran 1980). Significant differences among treatment means were calculated as per DMRT test (Duncan 1955).

RESULTS AND DISCUSSION

Per cent hatchability: Per cent hatchability on fertile egg set basis was higher in group where breeder vaccination and *in ovo* vaccination was performed followed by either non vaccinated or only *in ovo* vaccinated group (Table 1). *In ovo* vaccinated groups had higher hatchability compared to the groups in which no *in ovo* vaccination was carried out. There was no significant effect of maternal vaccination and *in ovo* vaccination on poult weight. This result concluded that *in ovo* vaccination did not affect the hatchability rather it improved the hatchability. Mast and Goddeeris (1999) reported that no major differences were observed in the hatchability and growth of broiler chickens immunized at ED16, ED18 and at hatch. Sharma and Burmester (1982) also reported that *in ovo* vaccination of Marek's disease (MD) did not affect the hatchability. However, Sudhagar (2006) reported that *in ovo* vaccination of live NDV vaccine reduced the hatchability (52.98%). It is to mention that he had used live NDV vaccine, whereas in the present study, the ND vaccine was modified by use of formaldehyde, which killed the virus thereby reducing virus proliferation and subsequent viraemic condition in embryo.

Hemagglutination inhibition titer: Haemagglutination inhibition titer (log₂) against ND vaccination was significantly higher (P<0.05) in poults hatched from maternally vaccinated hens than the un-vaccinated ones up to 28 days of age. Further, the poults, which received *in ovo* vaccination, had significantly higher titer than un-*in ovo* vaccinated poults till 28 days of age. Sudhagar (2006) reported that the peak titre appeared on fourth day post hatch

Table 1. Effect of maternal vaccination (MV) and *in ovo* (IV) vaccination on hatchability indices and poult weight

Group	Death before pipping (%)	Death after pipping (%)	Hatchability (%)	Poult weight (g)
MV + IV	16.39	0	83.61	52.33
MV + No IV	26.37	0	73.63	52.08
No MV +IV	18.24	2.70	79.05	52.57
No MV+ No IV	19.19	1.01	79.80	52.10
MV				
Vaccination	21.38	0	78.62	52.23
No vaccination	18.72	1.86	79.43	52.38
IV				
Vaccination	17.32	1.35	81.33	52.46
No vaccination	22.78	0.51	76.71	52.09
Pooled SEM	2.19	0.64	2.06	0.23
Sig level				
MV	NS	NS	NS	NS
IV	NS	NS	NS	NS
MV × IV	NS	NS	NS	NS

Means bearing different superscripts in a column differ significantly (P<0.05).

Table 2. Interaction of maternal vaccination (MV) and *in ovo* vaccination (IV), *in ovo* vaccination (IV) and conventional vaccination (CV), maternal vaccination (MV) and conventional vaccination (CV) on ND HI titre at 0, 7, 14, 21, 28, 42 and 49 days of age (d)

Groups	0d	7d	14d	21d	28d	42d	49d
MV×IV							
MV+IV	7.4 ^{ab}	9.25 ^b	9.08	8	8.33 ^b	6.17	6.38
MV+No IV	8 ^b	9.92 ^b	8.92	7.75	7.73 ^{ab}	6.5	5.92
No MV+IV	9.4 ^c	8.75 ^b	8.67	7.25	8.92 ^b	6.54	6.86
No MV+No IV	6.5 ^a	7.17 ^a	6.75	5.83	6.63 ^a	5.83	5.58
IV×CV							
IV+CV	8.4	8.67 ^{ab}	8.83	7.5	8.92	6.08	6.18 ^b
IV+No CV	8.4	9.33 ^b	8.92	7.75	8.33	6.63	7 ^b
No IV+CV	7.33	9.5 ^b	8.42	7.25	7.33	6.83	7.42 ^b
No IV+No CV	7.33	7.58 ^a	7.25	6.33	6.95	5.5	4.08 ^a
MV×CV							
MV+CV	7.7	10.17	9	8.25	7.75 ^{ab}	6.33	6.67
MV+No CV	7.7	9	9	7.5	8.36 ^{ab}	6.33	5.63
No MV+CV	8.11	8	8.25	6.5	8.5 ^b	6.58	7
No MV+No CV	8.11	7.92	7.17	6.58	7.04 ^a	5.79	5.46
Pooled SEM	0.27	0.27	0.30	0.26	0.24	0.21	0.33
Sig Level							
MV×IV	P<0.01	P<0.05	NS	NS	P<0.01	NS	NS
IV×CV	NS	P<0.01	NS	NS	NS	NS	P<0.01
MV×CV	NS	NS	NS	NS	P<0.002	NS	NS

Means bearing different superscripts in a column differ significantly (P<0.05).

and on seventh day post hatch in chicks vaccinated on ED15 and ED18, respectively, and also reported the interval between *in ovo* vaccination and the peak titre was 10 days in both cases. This is in line with the present finding where the peak HI titre was noticed on seventh day post-hatch in *in ovo* vaccinated group and it was 10 days after embryonic vaccination against NDV. Further, *in ovo* vaccinated poult had significantly higher titer at 0 (P<0.05), 14 (P<0.01) and 28 day (P<0.01) and apparently higher titer than un-*in ovo* vaccinated poult at 7, 21, 42 and 49 day. In other words, *in ovo* vaccinated poult had significantly (P<0.05) higher titer at 3 days, 17 days and 31 days after *in ovo* vaccination. Sudhagar (2006) reported higher titre was maintained up to 20 days after *in ovo* vaccination in 18th ED.

Combination of maternal vaccination and *in ovo* vaccination had higher titre value than only *in ovo* vaccinated group till 21 days of age. Thereafter only *in ovo* vaccinated groups had higher titre than either maternal or combination of both. However, maternal vaccination, *in ovo* vaccination or combination of both had higher titer values than unvaccinated poult.

In ovo vaccination also had apparently higher titer values than conventional vaccination till 28 days of age. This is in line with the findings of Bakyaraj (2007) who also reported that in conventionally vaccinated birds, the titre was lower than *in ovo* vaccinated. However, combination of *in ovo* vaccination and conventional vaccination did not show any beneficial effect on HI titer. Sudhagar (2010) also reported that chicks vaccinated on 18th ED maintained comparatively

higher titre than conventionally vaccinated chicks from 10 day post hatch and by 28th day the difference was significant. Conventionally vaccinated poult had apparently higher titre than the non vaccinated poult throughout the experiment. Peak titre was obtained on seventh day and titre was maintained by vaccinating at day-old and then again booster vaccination after 4 weeks of age. Neelima (1995) reported that by conventional vaccination, peak titre value was obtained on tenth day and high titre was maintained till 20 days post vaccination. However, combination of *in ovo* vaccination and conventional vaccination did not show any beneficial effect on HI titer.

Interaction of maternal vaccination and conventional vaccination on ND HI titre showed that only maternal vaccination maintained higher titer value than conventional vaccination up to 21 days of age. Thereafter, conventional vaccinated group had higher titer value. Combination of maternal vaccination and conventional vaccination did not elicit better immune response than either maternal or conventional vaccination. An *in ovo* vaccination, intra yolk sac (IYS) delivery of viral vaccines was attempted earlier. It was opined that as the yolk is absorbed over 5 to 7 days, any antigen introduced into the yolk sac might be released into the blood stream over that period. However, there has always been a concern that this type of *in ovo* vaccination may lead to interference with the maternal antibodies. Hence, in the present study, 25 mm needle length was used for *in ovo* vaccination and the virus was deposited mostly in amniotic sac.

The study envisaged that maternal vaccination along with *in ovo* vaccination for ND may maintain higher titre values till 49 days of age. Further, combination of maternal vaccination and conventional vaccination may not elicit better protective immunity against ND than those practiced alone.

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