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308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorijps@gmail.com

Experimental Vaccination Against Newcastle Disease in Japanese Quails (*Coturnix coturnix japonica*): Clinical and Immunological Parameters

Antonio Carlos Paulillo^{1,2}, Elizabeth Moreira dos Santos Schmidt^{1,3,4},

Janine Denadai⁵, Fabiana Silva Lima⁶ and Luciano Doretto Junior¹

¹Departamento de Patologia Veterinária, FCAV-Unesp, Jaboticabal, Brazil

²Research Fellow-CNPq, Brazil

³Post-doctorate Researcher in Veterinary Medicine of FCAV-Unesp, Jaboticabal, Brazil

⁴Bolsista FAPESP-Brazil

⁵Aluna Curso de Pós-graduação em Medicina Veterinária-FCAV, Unesp, Jaboticabal, Brazil

⁶Ministério da Agricultura, Pecuária e Abastecimento (MAPA)-Brasília, DF, Brazil

Abstract: Clinical and immunological parameters of vaccinated Japanese quails against Newcastle disease were evaluated. Two-hundred and forty birds were distributed into five different experimental groups, vaccinated or not against Newcastle Disease (ND): G1 (Ulster 2C strain), G2 (B1 strain), G3 (LaSota strain), G4 (LaSota strain inactivated and emulsified in mineral oil) and G5 (not vaccinated-control). The immune response was evaluated by the HI test. The vaccinations of Japanese quails with NDV LaSota strain inactivated and emulsified in mineral oil strain produced high antibody levels. Ulster 2C, B1 and LaSota live strains produced moderated antibody levels and did not cause any clinical signs associated with post-vaccinal reactions.

Key words: Japanese quail, Newcastle disease, *Coturnix coturnix japonica*, Ulster 2C, B1 and LaSota strains, vaccination

INTRODUCTION

Newcastle Disease (ND), caused by *Avian Parainfluenzavirus* serotype 1 (APMV-1) viruses, which is a member of the genus *Avulavirus*, of the *Paramyxoviridae* family (ICTV, 2007) is included in List A of the Office International des Epizooties. Historically, ND has been a devastating disease of poultry and in many countries the disease remains one of the major problems affecting existing or developing poultry industries. Even in countries where ND may be considered to be controlled, an economic burden is still associated with vaccination and/or maintaining strict biosecurity measures. The variable nature of Newcastle disease virus strains in terms of virulence for poultry and the different susceptibilities of the different species of birds mean that for control and trade purposes, ND requires careful definition. Control of ND is by prevention of introduction and spread, good biosecurity practices and/or vaccination (Kaleta and Baldauf, 1988; Alexander, 2000).

The commercial production of Japanese quails is extensively distributed in several countries around the world and many studies showed that this species can easily adapt to commercial management conditions, with good performance in terms of meat and egg production (Murikami, 1991). However, there is little information available on health control programs in this species. Thus, the present study aimed to evaluate vaccination programs against Newcastle disease in Japanese quails.

MATERIALS AND METHODS

Experimental birds and management: About 240 Japanese quails from 1-25 weeks of life were distributed in a completely randomized experimental design with 5 different treatments, with six replicates of 8 birds each. During the pre-experimental period (0-5 weeks of age), Japanese quails were vaccinated against Newcastle disease, except those in the control group, with Ulster 2C strain at 10 and 22 days of age by eye drop.

Japanese quails were allocated in experimental floor-pen housed, receiving water and food *ad libitum*. The feed was formulated with corn and soybean according to NRC (1994) recommendations.

Vaccines: Commercial line NDV vaccines (Ulster 2C, B1 and LaSota strains) were administered to each experimental group, as described by Paulillo *et al.* (1996). All birds, except those in the control group, were vaccinated at 5 weeks of age and revaccinated at 13 and 21 weeks of age with the same vaccine strain that was applied in the first vaccination. Vaccine titers were obtained by determining 50% of the embryo-infecting dose in embryonated eggs of specific-pathogen-free breeders at 8 and 10 days of incubation. Titers of live vaccine strains Ulster 2C, B1 and LaSota were 7.15, 7.2 and 7.35 log₁₀/0.1mL, respectively. Titer of the inactivated vaccine with LaSota strain was 9.5 log₁₀/0.1mL and this vaccine was emulsified in mineral oil. Birds were vaccinated and revaccinated by

Table 1: Mean antibody titres measured by HI test (\log_2) of Japanese Quails (*Coturnix coturnix japonica*) submitted to different vaccination programs against ND

Group	Vaccine	Japanese quails ages (weeks)							
		1	5	8	12	16	19	23	25
I*	Ulster 2C	0.0	0.0	4.2 ^c	3.4 ^b	4.4 ^a	3.6 ^a	5.6 ^a	4.8 ^a
II*	B1	0.0	0.0	4.6b ^c	4.2 ^b	3.8 ^a	4.2 ^a	6.3 ^a	4.8 ^a
III*	LaSota	0.0	0.0	6.0 ^b	5.0 ^{ab}	5.2 ^a	4.4 ^a	5.2 ^a	5.4 ^a
IV*	LaSota (oil)	0.0	0.0	8.6 ^a	7.0 ^a	6.0 ^a	4.2 ^a	0.0 ^b	0.0 ^b
V**	Control	0.0	0.0	0.0 ^d	0.0 ^c	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b

*Groups were also vaccinated with Ulster 2C strain at 10 and 22 days of age. **Control group-not vaccinated against ND. 1-Means followed by the same letter, in the same column, are not different at 5% of probability by Tukey test ($p>0.05$)

eye drop (Ulster 2C, B1 and LaSota-G1-G4) and vaccinated subcutaneously (LaSota inactivated-G5).

Serology: Blood samples of Japanese quails were collected from the jugular and ulnar superficial vein, from 1-25 weeks of age, at regular 3-4 week intervals. Sera were inactivated at 56°C for 30 min, frozen and stored at -20°C. Sera samples were submitted to inhibition of Hemagglutination (HI) test, according to Cunningham (1971).

RESULTS AND DISCUSSION

Mean antibody titres against ND from Japanese quails are shown in Table 1. Until 5 weeks of age, none of the birds showed maternally-derived antibodies against ND, as breeders were not submitted to any vaccination programs against this disease. As the control group (G5) was not vaccinated, its antibody titres were null during all the experimental period. Japanese quails from all groups vaccinated or not against ND did not show any clinical signs of post-vaccinal reactions.

The Tukey test demonstrated significant differences ($p<0.05$) between LaSota (inactivated) vaccinated group and the other vaccinated groups (Ulster 2C, B1 and LaSota activated), especially at 8 weeks of age.

At 8 weeks of age, antibody titres against ND were detected in the LaSota (inactivated) vaccinated group. These antibody titres were high and remained elevated until 16 weeks of age. The pre-experimental vaccination with Ulster 2C strain might have contributed to explain the immune status of the Japanese quails, with high antibody levels detected on the LaSota (inactivated) vaccinated group (G4). These high antibody titres detected for the Japanese quails vaccinated with LaSota strain (inactivated) (G4) are compatible with the great diffusion potential of this strain (Winterfield *et al.*, 1957). The oil adjuvant make a stable emulsion in which the antigen is slowly released, thus a prolonged immune stimulus is observed (Warden *et al.*, 1975). These results are similar to those reported by Paulillo (1988) in hens and in partridges (Paulillo *et al.*, 2008a) vaccinated with inactivated LaSota strain. However, Guinea fowls vaccinated with inactivated LaSota strain showed low to moderated antibody titres (Paulillo *et al.*, 2008b).

At 8 weeks of age, antibody titres against ND were detected in the vaccinated groups. This active immunity was induced by vaccination at 5 weeks of age. Japanese quails were revaccinated at 13 and 21 weeks of age and this procedure maintained antibody titres against ND up to 25 weeks of age. The low diffusion potential of the Ulster 2C strain (McFerran and Nelson, 1971) and the low invasion capacity of the B1 strain (Hofstad, 1951) may explain the low to moderated antibody titres detected by HI in vaccinated Japanese quails. On the other hand, the low antibody titres detected for the Japanese quails vaccinated with the LaSota strain (activated) (G3) are not compatible with the great diffusion potential of this strain (Winterfield *et al.*, 1957). The analysis of these serological results clearly shows that Japanese quails produce antibody when vaccinated against ND.

Conclusion: Our study has shown that commercially available ND LaSota strain inactivated vaccine for chickens induced a high antibody response in Japanese quails. The present study also showed that Japanese quails produced a moderated antibody response when vaccinated with commercially available live vaccines (Ulster 2C, B1 and LaSota) for chickens against Newcastle disease, without any clinical signs of post-vaccinal reactions.

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