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A Comparative Study of Levamisole and Dexamethasone on the Chicks Immunity Which Inoculated with Newcastle Vaccine

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Abstract: The objective of the study reported here was to evaluate the effects of Levamisole and dexamethasone on antibody titer in chicks against Newcastle vaccine.120 Fertile eggs at 18 days of incubation were divided into four equal group, 30 embryonated egg for each. The first group A inoculated with Levamisole that dose 0.1 mL/egg contain 1.25 mg active ingredient and live attenuated Newcastle vaccine(Lasota strain) in dose 0.1 mL/egg contain $4x10^8$ Antigen in chick embryo at 18 days. The second group B injected with Dexamethasone that dose 0.1 mL/egg contain 0.2 mg/egg and Newcastle vaccine (Lasota strain) in dose 0.1 mL/egg contain $4x10^8$ Antigen, in chick embryo at 18 days. The third group C treated with live attenuated Newcastle vaccine (Lasota strain) alone and saved as positive control in chick embryo at 18 days, whereas forth group D (negative control) injected with normal saline in dose 0.1 mL/egg at the same age and method of in group D. Result showed that slight effect of levamisol in increase Hatchability and livability ratio and it indicated sever effect of Dexamethasone in decreased Hatchability and livability. Heamagglutination inhibition test (HI) was used to evaluate the antibodies titer for all groups at 14 days of age. The above results showed the slight increase in HI titer for group A but it was not statisically significant as compared with group C, whereas result of group B showed significant decrease in HI titer as compared with group C.

Key words: Levamisole, dexamethasone, antibody titer,

INTRODUCTION

Newcastle Disease (ND) is a contagious, highly fatal viral infection affecting many species of domestic and wild birds worldwide. This is due to the huge economic impact on poultry industry precipitated following outbreaks of the disease (Aldous and Alexander, 2001).Traditionally, synthetic chemicals and antibiotics have been used to prevent or treat poultry diseases and have achieved at least partial success. However, the emergence of antibiotic-resistant microorganisms and antibiotic residues in meat are the most limiting matter to their expanded usage. Therefore, vaccination against specific pathogens has been developed with variable degrees of successes. Such successes depend on the particular factors such as special antigens, immunogenicity of antigens and immune stimulants. Use of immune stimulants for the prevention of diseases in poultry is considered an effective and improving area. Immuno stimulants are natural or synthetic substances able to enhance the non-specific and the specific immune responses (Anderson, 1992). Levamisole (LMS) is a synthetic antihelmintic drug for animals against stomach, intestinal and lungworms (Janssen, 1976). LMS is a promising agent for use in the immunotherapy of patients with deficient host defense mechanisms. LMS has been shown to stimulate cell mediated immunity probably through the enhanced maturation of cells (Sampson and Lui, 1976). Dexamethasone (DEX), often regarded as a hallmark of stress, play a critical role in affecting physiological and immunological changes, such as anemia, body weight loss, increased body temperature and respiratory rate and reduced growth rate in stressed animal (Dohms and Metz, 1991). Age of exposure to DEX appears to be an important factor in immune outcome. In poultry, however, data on the use of DEX are scarce special in the hatching stage (Coe *et al*., 1999).

The purpose of this study was to evaluated the effect of LMS as immune stimulator and used DEX as immune suppressor on antibody titer using laSota attenuated vaccine used in embryo.

MATERIALS AND METHODS

A total of 120 Fertile eggs were obtained from local commercial hatchery. The eggs were randomly divided in to four groups A,B,C and D. Each group containing 30 eggs. Then the eggs were incubated by used incubator until 18th days and candling has been reduced the number of eggs.

At 18th days the experimental groups were treated, group A was performed for testing the effect of LMS on immune status, this group received by inoculation 0.1 mL of LMS which contained 1.25 mg/egg and then inoculated with 0.1 mL of laSota attenuated vaccine which contain $4x10^8$/egg. Group B were conducted to explain influence of DEX as immune suppressor on immunity, this step done by inoculated 0.1 mL of DEX which containing 0.2 mg/egg and then the eggs inoculated with laSota attenuated vaccine which contain $4x10^8$/egg. Group C were inoculated with 0.1 mL laSota

Newcastle vaccine which contain $4x10^8$/egg and regarded as positive control where as group D received 0.1 mL normal slain were considered as negative control. The eggs were then transferred in to incubator and they remain until hatch at 21st days.

The incubation method which has been used according to (Ahmed and Sharma, 1992). The inoculation method done by used disposable medical syringes capacity 1ml with needle 2.5cm and diameter 22G (Stone *et al*., 1997).

Blood sample from 10 randomly selected birds of each group were collected on 14th days of age and serum was separated for estimation of antibody titer against ND vaccine. After collection serum sample were stored at -20°C until used hem agglutination inhibition test (HI) testing. Prior to conducting HI, the serum samples were thawed. Phosphate buffer saline (PBS) solution having a PH of 7.2 was used in hem agglutination test (HA) and HI tests. Erythrocyte was collected from ND antibody free chickens, blood were washed and their 1 percent solution was used in HA and HI tests. The HA and HI tests were conducted according to the protocol described by (Allan and Gangh, 1974). Hatchability was calculated by, number of hatched birds/number of fertile eggs x 100 (Sharma and Burmester, 1982). The livability was carried out according to the well established principles and protocol (Ahmed and Sharma, 1992) by number of live birds/number of dead birds x 100. The data were subjected to analysis of variance and the significant differences at ($p<0.05$) were determined by ANOVA-one way (SPSSv.12,2004).

RESULTS AND DISCUSSION

In the present study results of exposure hatching eggs to Levamisole and dexamethasone at 18th day of incubation showed in Table 1.

Table revealed lower hatchability (73%) and low livability (73.3%) in group B which inoculated with (DEX and ND vaccine). These result was statistically significant as compared with other groups especially group C. Decreasing of hatchability and low livability in group B may be attributed to the harmful effect of DEX on embryos. These results were in agreement with that reported by Rombauts *et al*. (1994) who found that DEX has stress show on hatchability and immunity. Also these results were in line with previous studies such as study of Allan and Gangh (1974); Stevan *et al*. (2001) and Huff *et al*. (1999). They mentioned that treat chicken with DEX increased mortality and it is act as Inhibitor factor for T-cells growth factor and B-cells growth, this due to decreased immune status and hatchability.

The hatchability and livability of group A were (93 and 100%), respectively, the both results of treatment had a minor increased than that of control group C. Statistically there was no difference between this group and group C. These results demonstrated that treatment with LMS at 18th day, didn't appear to have an influence on hatchability and livability.

Table 1: Effect of treatment on incubated eggs at 18th day of incubation on hatchability and Livability

Groups	No.of hatched bird/No. of fertile eggs	H	No. of live bird/no. of dead bird	L
A (LMS+ND vaccine)	28/30	93	28/0	100
B (DEX+ND vaccine)	22/30	73	22/3	73.3
C (ND vaccine)	27/30	90	27/1	96
D (Normal slain)	27/30	90	27/0	100

H: Hatchability, L: Livability

Table 2: Effect of treated on HI titer after hatching at 14th day

Groups	Type of treat	No. of sample	Mean of HI titer (log_2)±SE
A	LMS+ND vaccine	10	$6.50±0.166^a$
B	DEX+ND vaccine	10	$5.00±0.333^b$
C	ND vaccine	10	$6.00±0.258^a$
D	Normal slain	10	$2.00±0.331^c$

SE: Standard Error

a,b,c and d figures with different superscripts in the vertical column were significantly different at ($p<0.05$)

These results were in agreement with Pioter *et al*. (2003) who reported that there was no effect of LMS has been established on hatchability and level of embryo mortality during the period of embryos development.

On other hand result of group C showed that vaccination used in the injection of embryo had no effect on hatchability and livability rations. These results were in line with Stone *et al*. (1997) who published that vaccination at 18th days of incubation had not affecting on hatchability and livability.

The antibody titer as detected by HI test in all groups are demonstrated in Table 2.

The result presented in the table indicated that antibody titer of chicks in group A are increased compared with other groups. Although numerical increased in HI has been shown for group A but this result is not statistically significant compared with positive control group C. This result was in agreement with that of jin *et al*. (2004) and Kang *et al*. (2005) who demonstrated that LMS stimulates T-cell activation and increased the production of antibody, using co- administered LMS. Also Cuesta *et al*. (2002) published that, LMS has been reported to increased antibody response against LaSota antigen in chicks.

Kulkarni *et al*. (1973) reported that chickens immunized with ND virus developed a higher level of HI antibodies when treated with LMS than untreated ones. In previous study by Yin *et al*. (2006) and Chawak *et al*. (1993) they observed that the LMS can enhance lymphocyte proliferation and it is act as a multifunctional modulator after immunization to mediated the cell-mediated response of T-cells and the same time promote

activation B- cells to produce antibody, this is another possible method of LMS to stimulate immune system.

The mean of HI test of ND vaccine in group B was (5.00), this result was statistically significant with other groups especially group A and C. Decreasing of immune response in group B attributed to immune suppressive of DEX. This result was in agreement with that of Dowling (1998) who reported that DEX inhibits the release of inflammatory mediators from macrophage and eosinophils, also decreased synthesis of prostaglandins leukotrienes and platelets-activation factor which play important roles in the suppress immune response.

Ogunsanmi *et al*. (1994) mentioned that DEX suppress both inflammatory and immunological response and inhibition a number of lymphocyte. Also Davison *et al*. (2006) indicated that treat chickens with DEX inhibit interleukin.

Result of group C was (6.00) statistically significant compared with group D, this is increased in HI titer attributed to effect of vaccine at 18th day of incubation. These result was in accordance with that of Ahmed and Sharma (1992) who found that ND vaccine may be used as embryo to protect chickens against ND disease.

Group D was (2.00) indicated a low HI titer at 14th day compared with other group. These findings strongly support the findings of Balla (1986); Saeed *et al*. (1988) who stated that the persistence of MDA in chickens were day 15 to 20 of age. Also Alexander (2003) who published that a decreased in maternal antibodies gradually until 14-21st days of age.

But Mahmud *et al*. (2007) reported that the persistence of MDA upto 27 days of age which may be due to the high MDA titres.

Conclusion:

1. Slight effect was observed on hatchability, livability and HI titer, after treat hatching eggs at 18 days of incubation with LMS
2. Oculation DEX in chicks embryo at 18 days of incubation resulted decreasing hatchability, livability and HI titer against ND vaccine

REFERENCES

Ahmed, J. and J.M. Sharma, 1992. Evaluation of modified live virus vaccine administered in ovo to protect chickens against Newcastle. Am. J. Vet. Res., 53: 1999-2004.

Aldous, E.W. and D.J. Alexander, 2001. Technical review.Detecation and differentiation of Newcastle disease virus (avain paramyxo virus type 1). Avian Patho., 30: 117-129.

Alexander, D.J., 2003. Newcastle disease and other paramyoxvirus. Pneumovirus. In: Disease of poultry, 11th Ed., Eds. by Calnek, B.W., Saif, Y.M., Mcdougald, L.R. and Swayne, D.E. Iowa state press, pp: 64-81.

Allan, W.H. and R.E. Gangh, 1974. A standard hemagglutination inhibition test for Newcastle disease. Vet. Res., 195: 120-123.

Anderson, D.P., 1992. Immunostimulants, adjuvants and vaccine carriers in fish: applications to aquaculture. Ann. Rev. Fish Dis., 2: 281-307.

Balla, L., 1986. Use of a standardized HI test for monitoring immunity to Newcastle disease. I. Experiments to standardize the HI test II. Antibody response after different immunization schedules. Magyar Allatorvoso Lapja, 41: 98-109.

Chawak, M., B. Rajmane and A. Rande, 1993. Effect of levamisole on performance and immunomodulation against Ranikhet disease in broilers under stress. Ind. J. Anim. Sci., 63: 1060-1061.

Coe, C.L., G.R. Lubacha and J.W. Karaszewski, 1999. Prenatal stress and immune recognition of self and nonself in the primate neonate. Biol. Neonate, 76: 301-310.

Cuesta, A., M. Esteban and J. Meseguer, 2002. Levamisole is a potent enhancer of gilthead seabream natural cytotoxic activity. Vet. Immunol. Immunopathol., 89: 169-174.

Davison, T.F., T.R. Morris and L.N. Payne, 2006. Poultry Immunology. 1st. Ed. Curfax Publishing company. U.K.

Dohms, J.E. and A. Metz, 1991. Stress mechanisms of immunosuppression. Vet. Immunol. Immunopathol. 30: 89-109.

Dowling, P.M., 1998. Glucocorticoids. In: The Merck Veterinary Manual, 8th ed. (Aiello, S.E., ed.), Merck and Co., Inc., New Jersey, U.S.A., pp: 1729.

Huff, G.R., W.E. Huff, J.M. Balog and N.C. Rath, 1999. The Effects of Dexamethasone Immuno-suppression on Turkey Osteomyelitis Complex in an Experimental *Escherichia coli* Respiratory Infection. Poult. Sci., 77: 654-661.

Janssen, P.A.J., 1976. The Levamisole story. Prod. Drug Res., 20: 347-383.

Jin, H., Y. Li, Z. Ma, F. Zhang, Q. Xie, D. Gu and B. Wang, 2004. Effect of chemical adjuvants on DNA vaccination. Vaccine, 22: 2925-2935.

Kang, Y., H. Jin, G. Zheng, Q. Xie, J. Yin, Y. Yu, C. Xiao, X. Zhang, A. Chen and B. Wang, 2005. The adjuvant effect of levamisole on killed viral vaccines. Vaccine, 23: 5543-5550.

Kulkarni, V., A. Mulbagal, V. Paranjape, J. Khot and A. Manda, 1973. Immunostimulating effect of tetramisole on antibody formation against Newcastle disease virus in chicks. Ind. Vet. J., 50: 225-227.

Mahmud, M.S., M.T. Hossain, P. Monoura and M.M. Amin, 2007. Comparative efficacy of Avinew (VG/GA strain) and BCRDV (F strain) vaccines against Newcastle disease in broiler chickens. Bang. J. Vet. Med., 5: 19-23.

Ogunsanmi, O.A., S.O. Akpavie and V.O. Anos, 1994. Haematological changes in ewes experimentally infected with *Trypanosoma brucei*. Rev. Elev. Med. Vet. Pays. Trop., 47: 53-57.

Pioter, S., k. Ewa, B. Wojciech, B. Wanda and K. Grazyna, 2003. Evaluation of chosen Immuno Wodualters Toxicity for chicken Embryos in one day old chicks. Bull. Vet. Inst. Pulawy, 47: 411-441.

Rombauts, L., D. Vanmont, Fort, L.R. Berghman and E. Decuprtr, 1994. Contribution of the Fetal adrenal to circulating Immune active inhibin in the chiken embryo. Biol. Ropert, 51: 926-933.

Saeed, Z., S. Ahmad, A.R. Rizvi and M. Ajmal, 1988. Role of maternal antibody in determination of an effective Newcastle disease vaccination programme. Pak. J. Vet. Res., 1: 18-21.

Sampson, D. and A. Lui, 1976. The effect of levamisole on cell-mediated immunity and suppressor cell function. Cancer Res., 36: 952.

Sharma, J.M. and B.R. Burmester, 1982. Resistance to Mareks disease at hatching in chickens vaccinated as embryos with turkey herpes virus. Avian Dis., 26: 134-149.

Stevan, G., R. Gerald and A.S. Grabteer, 2001. Challenge model for Adult nor then bob white (colinus virg imianus) using vaccine strain of *Pasteurella multocida* type 3. Avian. Dis., 40: 99-102.

Stone, H., B. Mitchell and M. Burgh, 1997. In ovo vaccination of chicken embryo with experimental Newcastle disease and avian influenza oil emulsion vaccines. Avian Dis., 14: 856-863.

Yin, J., H. Jin, Y. Kang, C. Xiao, L. Zhao, X. Li,Z. Ding, F. Yang, Q. Zhu and B. Wang, 2006. Efficacy of modified levamisole adjuvant on inactivated virus vaccine.Viral Immunol., 19: 525-535.