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# Effects of different levels of zinc supplementation on broilers performance and immunity response to Newcastle disease vaccine

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# ABSTRACT

Zinc is a nutritionally essential trace element that acts in most metabolic pathways. Nowadays the use of specific dietary supplements to boost the intrinsic potential of poultry to perform better immunologically is so important. The aim of the present study was to investigate the zinc additive different levels effect on immune response and performance of broilers. In this study, 200 Ross 308 broilers divided in 5 groups. In group1, 2, 3 and 4: 50, 75, 100 and 125 ppm Zinc, respectively, were used for two weeks. In group5: was mentioned as a control group. This study was performed in 42 days period and growth parameters include body weight gain, feed intake and FCR were calculated in all groups and antibody titers against Newcastle disease were evaluated by HI and ELISA methods. Dietary Zn significantly affected daily body weight gain and daily feed intake (p<0.01) but not the feed conversion ratio, and the best results were obtained in 100 ppm groups. Dietary Zn significantly affected antibody titers against Newcastle disease (p<0.01), and the highest antibody titer in ELISA and HI methods was in 100 ppm groups. Our results indicated that 100 ppm Zn supplementation could improve performance of broilers also it has immunestimulatory effects and increase humoral immunity response to vaccination.

Keywords: Zinc, broilers, performance, immunity, Antibody titer

# INTRODUCTION

Zinc (Zn) is a nutritionally essential trace element that acts in most metabolic pathways. It plays a critical role in immune function as moderate Zn deficiency and severe Zn deficiency are known to affect immunity in human and animal models [9, 16, 17, 36]. The most commonly used Zn for supplementation in animal diet is inorganic Zn in the form of zinc sulfate due to cost and commercial availability. Nowadays the use of specific dietary supplements to boost the intrinsic potential of poultry to perform better immunologically is so important. Trace elements are involved in the metabolic activities via metalloenzymes which are essential for the antioxidant protection of cells in poultry [32].

Zinc is essential for highly proliferating cells, especially in the immune system and is an essential cofactor for thymulin which modulates cytokine release and induces proliferation [26]. There are conflicting results regarding the level of zinc required to affect an immune response. Some studies indicate that supplementing the diet of broilers above the National Research Council (NRC, 1994) recommendation could enhances antibody production [19], whereas the others have reported no effect [33].

Bartlett and Smith (2003) reported the broilers receiving 68 and 181 mg Zn had a higher response for total, IgM, and IgG antibodies [1].

Studies on the relationships between animal nutrition and immunity have sought reliable methodologies to measure responses. It is clear that zinc affects multiple aspects of immune system, from the barrier of the skin to gene regulation within lymphocytes. Zinc is crucial for normal development and function of cells mediating nonspecific immunity such as neutrophils and natural killer cells [36].

Dietary Zn might also influence the immune system indirectly by interaction with growth and infectivity of organisms that are pathogens to animals. Chickens have hypozincemia when infected with the Newcastle disease virus [38] or *Escherichia coli* endotoxin [5]. Zinc concentrations in the liver are increased by *E. coli* endotoxin infection [23]. Temporal and quantitative changes in zinc concentrations in immune tissues might be important in the response to infection because the host uses zinc as a cofactor for enzymes involved in defense against pathogens [23, 31]. Increased liver weight by infection could be the result of interleukin (IL-1) stimulation of acute-phase reactant protein synthesis such as metallothionein and ceruloplasmin [31]. Zn concentrations in serum and plasma are initially depressed when birds are infected with *Salmonella gallinarum* [13], *E. coli* [39], or *E. coli* endotoxin [5].

The effectiveness of Zn inhibition of bacterial growth results from changing the active transport system and impeding the initial phase of bacterial mating [37]. They also propose that zinc treatments such as zinc chloride enhanced survival incidence in rats infected with *Francisella tularensis* or *Streptococcus pneumoniae*. This can be explained by zinc's association with defense mechanisms such as leukocytosis, phagocytosis, and cell-mediated immunity as well as indirectly by zinc's inhibition of proliferation of these foreign invaders.

Burrell et al. [4] reported improved performance when broilers consumed diets formulated to contain 110mg Zn/kg; while the NRC (1994) mentioned that the requirement of Zn for broiler chickens is only 40 mg/kg [29]. Excessive supplement of inorganic Zn can bring on serious environment pollution resulting from the low utilization of this element. Therefore, excretion of Zn may be reduced by using sources of Zn with higher availability. Zinc from amino acid complexes has been reported to be more bioavailable than Zn from inorganic sources [6, 42].

Zinc is an essential micro-mineral that is involved in several metabolic routes that are fundamental for growing and living[25]. This mineral is essential for the immunological function and disease prevention, since it improves the responses to antigenic challenges under field conditions [21]. Adequate Zn consumption is crucial to the development, maintenance and efficient functioning of the immunological system and the cells associated to it [8]. Nevertheless, previous studies have not proven that zinc has influence on chick immunological response [20, 34]. The objective of this study was to evaluate the zinc additives different levels effect on immune response and performance of broilers.

### MATERIALS AND METHODS

In this study, 200 Ross 308 broilers divided in 5 groups, and each groups divided to 4 replication with 10 birds in each of them. In group1, 2, 3 and 4: 50, 75, 100 and 125 ppm Zinc, respectively, were used for two weeks from day 10 to 24. In group 5: placebo was used and that group mentioned as a control group. This study was performed in 42 days period and growth parameters include body weight gain, feed intake and FCR were calculated in all groups. For evaluation of zinc effects on immune response to vaccination the Newcastle vaccines titer were investigated by HI and ELISA methods.

Newcastle disease vaccination programs include clone-bronchitis on 12 days old and clone30 on 25 and 38 days old by drinking water.

**Statistical Analysis,** For comparison results between groups the data obtained were compared by One-way Analysis of variances (ANOVA) at 95% probability and in case of significantly statistic difference in ANOVA results, Duncan test at alpha level 0.05 was performed.

#### RESULTS

#### **Growth Performance**

Dietary Zn significantly affected daily body weight gain and daily feed intake (p<0.01) but not the feed conversion ratio (Table 1). When the diet was supplemented with 100 ppm Zn, chick BW gain and feed intake was improved, although there was not significant differences between feed intake and BW between 100 and 125 ppm groups.

Group	Body weight (g/bird)	Feed intake (g/bird)	Feed conversion ratio (FI/BW)
Control	2680.24±11.54 <sup>a*</sup>	6030.53±57.73 <sup>b</sup>	2.25±0.02
50 ppm	2689.91±6.35 <sup>a</sup>	5969.58±39.83 <sup>b</sup>	2.22±0.05
75 ppm	2705.06±8.66ª	5997.61±49.10 <sup>b</sup>	2.19±0.04
100 ppm	2744.12±7.50 <sup>b</sup>	5817.28±38.68 <sup>a</sup>	2.12±0.02
125 ppm	2734.67±8.08 <sup>b</sup>	$58.78.10 \pm 42.14^{ab}$	2.15±42.01
P value	0.001	0.046	0.162

\* Mean values within the same variable with unlike superscript letters were significantly different (p < 0.05).

#### Antibody titers

Dietary Zn significantly affected antibody titers against Newcastle disease (p<0.01) (Table 2). When the diet was supplemented with 100 ppm Zn, HI titer in 100 ppm group was highest and it was significantly different from 50 and 75 ppm groups, but in view of ELISA titers there was not significant different between 75, 100 and 125 ppm groups. Also our results indicated that the Zn addition to broilers drinking water could improve antibody titers against Newcastle disease and improves immune response to vaccination.

Group	ELISA	HI
Control	9863.50±659.69 <sup>a*</sup>	6.06±0.30 <sup>b</sup>
50 ppm	10347.05±927.09 <sup>a</sup>	7.22±0.32 <sup>b</sup>
75 ppm	12967.72±746.84 <sup>b</sup>	7.20±0.33 <sup>b</sup>
100 ppm	14800.22±535.29 <sup>b</sup>	8.39±0.30 <sup>a</sup>
125 ppm	14426.72±401.70 <sup>b</sup>	$8.06 \pm 0.16^{ab}$
P value	0.001	0.001

\* Mean values within the same variable with unlike superscript letters were significantly different (p<0.05).

#### DISCUSSION

During study, there was significant difference in body weight gain and feed intake between the control and experimental groups (100 and 125 ppm groups, Table 1). But there was no significant difference (p>0.05) in FCR between experimental and control groups.

Researchers previously indicated that not more than 61 mg/kg of supplemental Zn was needed in addition to the 41 mg/ kg present in the basal diet [10]. Burrell et al. (2004) reported that optimum BW was achieved at 110 mg of supplemental Zn/kg with a maize-soybean meal basal diet [3]. Other researchers results purposed when a soybean meal diet was fed to chicks from 1 to 21 d post hatching, the Zn requirement is 71 mg/kg [14]. Edwards et al. (1958) and Burrell et al. (2004) used BW as the criterion for Zn requirement, but the difference between those studies was the type of chicks used [4, 10]. The estimate for Zn requirement varied depending upon which response criterion was used [43].

Differences in criteria chosen and the differing type of chicks may offer explanations to why the requirement estimates differed among the experiments. The current NRC Zn requirement for chicks (40 mg of Zn/kg from 0 to 3 wk) was based on the level of Zn needed to maximize growth. However, growth may not be the best index of Zn status. When fed with purified or semi purified basal diets, optimal dietary Zn improves the early growth of broilers significantly [41, 44], but when fed with C-SBM diet, growth is usually not influenced [42]. The Zn in C-SBM basal diet often met the estimated requirement of the chick for Zn (40 mg/kg Zn; NRC, 1994).

Our result is similar to that reported by Mohanna and Nys (1999), who found that chick BW gain and food intake increased with the dietary Zn content until supplementation with 25 mg of Zn/kg was reached [28], and Salabi et al., (2011) results that they indicated the addition 90 mg/Kg total dietary Zn significantly improves body weight gain and feed intake. These data suggest that the requirement for early chick growth is satisfied when chicks are fed diets containing 40 mg of Zn/kg as recommended by the NRC (1994).

The immune system is dependent on the functions of cellular metabolism. Zinc is ubiquitous in cellular metabolism and functions both structurally and catalytically in metalloenzymes [30]. Other researchers have demonstrated that supplementation of broiler breeder hen diets with zinc promoted the cellular immune response of progeny [20, 22]. In addition, hens provided diets with added zinc from amino acid complexes or chelates had increased thymus weights [40] and improved livability of progeny[11, 35].

Hudson et al. (2004) reported that antibody titers to NDV were higher when supplemental zinc was provided by Znamino acid complex [15]. Similarly, in their study with broiler breeders, Khajarern et al. (2002) found that NDV, infectious bursal disease and infectious bronchitis titers were increased when hen diets were supplemented with zinc-amino acid complex [18]. Zinc is essential for thymulin, a thymic hormone that regulates T lymphocyte maturation [12]. Birds provided diets supplemented with a more available zinc source (ZnMet) might have induced thymulin activity, and therefore promoted immune responses through increased maturation of T-lymphocytes and activation of B-lymphocytes by T-helper cells. Consistent with our findings, Beach et al. (1980) reported that diets supplemented with zinc tended to improve the ability of birds to produce antibodies [2]. Also, Bartlett and Smith (2003) showed that birds receiving a high zinc diet had significantly higher titers of total, IgM and IgG antibodies than those receiving adequate or low zinc diets during the primary response [1]. Furthermore, those birds receiving adequate and high zinc diets were similar with higher responses for total, IgM, and IgG antibodies during the secondary challenge with SRBS than those in the low zinc group [1, 7]. Kidd et al. (1992) also reported that supplemental Zn from amino acid complexes in the diets of the parents and chicks increased antibody responses of chicks to SRBC and *Salmonella pullorum* challenges [20]. Also other researchers indicated that the dietary supplementation with organic zinc improves both cellular and humoral immune responses [24, 27].

Our result is similar to other researcher's results and Zn supplementation increased antibody titers against Newcastle disease.

## CONCLUSION

Our results indicated that 100 ppm Zn supplementation could improve performance of broilers include increase body weight gain and decrease feed intake, but it has not any effects on feed conversion ratio.

Also the results showed the antibodies obtained in Zn supplemented groups, especially in 100 ppm group were significantly higher than the other groups. With regards to our results Zn uses in broilers recommended.

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