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Gross and Microscopic Study of Different Organs in Zinc Intoxicated Male White Leg Horn

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ABSTRACT

The aim of this study was to evaluate the effects of excess zinc on the gross and microscopic changes of different organs along with performance parameters in male white leg horn (WLH) chicks. A total of 80 WLH male birds of 4 weeks age were randomly and equally divided into four groups (C, G1, G2 and G3). Zinc sulphate was incorporated in drinking water @ 15000 ppm, 25000 ppm and 35000 ppm to group G1, G2 and G3, respectively, for a period of 8 weeks. The birds from each group were sacrificed at the end of the experimental trial for pathomorphological changes and estimation of organ body weight ratio. Significant histopathological changes were noticed in liver, spleen, gizzard, bursa kidneys and pancreas. Atrophy of liver, testes, spleen, bursa, pancreas and gizzard were also noticed. This study confirms that, the high levels of zinc have an adverse effect on health and performance of the birds.

Keywords: Cockerels, Pathomorphology, Performance, toxicity, Zinc

Poultry industry contributes a major share in terms of protein supplement through meat and eggs in human food. The poultry industry which has earlier been termed as an unorganized sector has turned itself as an organized sector through people, process and technology. The major factors for successful poultry production are high genetic potential, balanced nutrition and health maintenance. The cost of feed accounts 65-70% for broiler and 75-80% for layer and is a major means of modulating the production cost. The feeding cost alone being the major constraint in poultry enterprise, every effort is directed to maintain feed quality, contain feed cost and utilize every bit of the feed to convert it into food for the mankind. Optimizing and updating nutrient requirements of poultry is a continuous process because of improved performance, variations in nutrient availability, interaction of different nutrients at sites of absorption and metabolism, and to achieve specialized designer products. The role of trace minerals in animal production is an area of interest for producers, feed manufacturers, veterinarians and scientists. Adequate intake and absorption of trace minerals is required for a variety of metabolic functions including growth, production, reproduction, skeletal development and maintenance of normal immune function. Trace mineral research has received less attention in the past and the recommendation on trace mineral requirements are mainly based on earlier research (Nutritional Research Council, 1994).

Zinc (Zn) is one of several micronutrients that have attracted increased attention because of its important role in maintaining physiological and metabolic functions such as physical growth, immuno-competence, reproductive function, and neurobehavioral development. Broilers and laying hens can tolerate 1-2 g kg^{-1}DM. Increases in Zn concentration (up to 4 g kg^{-1}DM) lead to loss of appetite and retarded growth (OH *et al.* 1979). Several reports have confirmed that the performance of broiler fowl is not adversely affected by the dietary inclusion of zinc oxide (ZnO) up to 1 g Zn/kg diet (Johnson *et al.* 1962; Kincaid *et al.* 1976). Although the supplementation of diets with excessive levels of Zn compounds has been suggested as a method for the induction of clutch in laying hens (Creger and Scott, 1977; Shippee *et al.* 1979). Hermayer *et al.* (1977) and Palafox *et al.* (1980) demonstrated that the dietary inclusion per kg of Zn compounds, at levels 10 and 20g leads to marked reduction in food consumption, egg production and body weight. Moreover Gentle *et al.* (1982) identified a threshold level of ZnO at or above 6 g Zn/kg diet that lead to reduction of feed intake of adult hens. Therefore, the present study was planned to study the effects of zinc at different but higher doses on performance and pathomorphological parameters in cockerels.

MATERIALS AND METHODS

The study was designed after getting approval from ethics committee. The experiment was carried out by using eighty white leghorn cockerels of one month old. After one week of acclimatization the birds were randomly divided into four groups viz control, G1, G2 and G3 with 20 birds in each. Zinc sulphate was administered via drinking water at the dose rate of 15000, 25000 and 35000 ppm to group G1, G2 and G3 respectively for a period of 8 weeks. Feed and water was provided in *adlibtum* throughout the study. At the end of experimental trial, postmortem of all the experimental groups was done and gross changes were recorded. Samples of liver, kidney, pancreas, bursa, gizzard and spleen were collected in 10% formalin for detailed pathomorphological studies. The birds from each experimental group were weighed fortnightly using weighing balance to study the effect of excess zinc administration on weight of the birds and were continuously observed for different clinical manifestations throughout the trial. Statistical analysis of the data was done by using ANOVA technique at 5% level of significance (Snedecor and Cochran, 1989).

RESULTS AND DISCUSSION

In the present study, the birds from group GI (15000 ppm) did not show any clinical signs during the entire period of study. While in birds of group G2 (25000 ppm) and G3 (35000 ppm) clinical signs of depression, weakness, emaciation, stunted growth and marked decrease in feed intake was observed throughout the study. The recorded mortality rate was 20% in group G3 (4/20). However no mortility was observed in G1and G2. Similar clinical signs were also observed by (Hermayer *et al.* 1977; Palafox *et al.* 1980) following dietary inclusion of zinc compounds @ 10g and 20g/kg feed in poultry. Gentle *et al.* (1982) reported rapid reduction of feed intake in adult hens following the inclusion of zinc oxide in the diet @ 6g/kg diet.

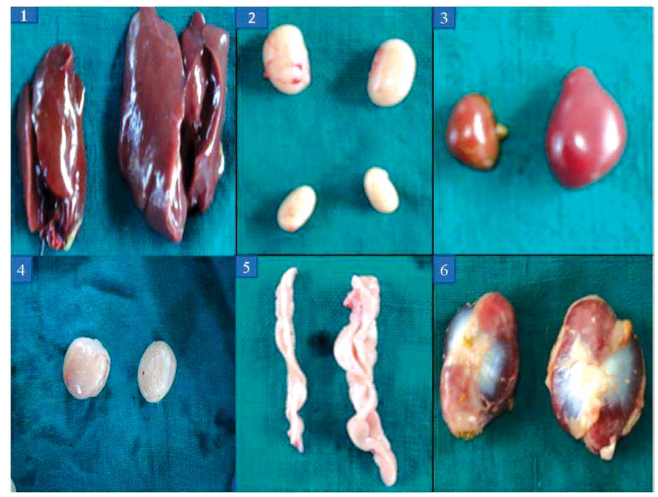

Fig. 1. Organs (1) Liver: shows atrophy and congestion in zinc intoxicated group with normal healthy control. While as in (2) testes (3) spleen (4) bursa of fabricius (5) pancreas and (6) gizzard, atrophy was observed in zinc intoxicated groups with normal healthy control

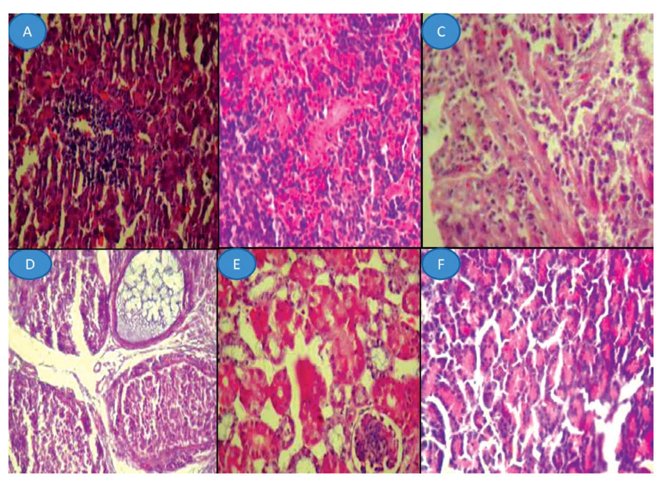

Fig. 2. (A) Liver showing hepatocytes at various stages of degeneration, distortion of hepatic cords and dilatation of sinusoids (400X). (B) Spleen reveals extensive haemorrhages (400X). (C) Koilin layer of gizzard showing tattered vertical rods with

infiltrated heterophils (400X). (D) Bursa of Fabricius showing cystic degenerative changes and depletion of lymphocytes in lymphoid follicles (400X). (E) Kidney showing extensive degeneration and necrosis of tubular epithelium and presence of solid dense coagulated mass of protoplasm in tubules (400X). (F) Pancreas showing dilatation of acinar lumina and dissorted acini (400X).

The average body weights recorded in the present study was in the descending order from control group to higher dosage groups. A significant decrease ($P<0.05$) in the body weight was recorded in group G3 followed by G2 and G1 as compared to control group (Table 1). Decrease in feed intake, malabsorption and interference of Zn with other dietary components and hepatic dysfunction may be the reasons for retarded growth observed in Zn intoxicated birds. These results were in agreement with the findings of (Hermayer *et al.* 1977 and Palafox *et al.* 1980) who also observed significant decrease in body weight gain in poultry with inclusion of high dietary zinc @ 10g and 20 g/ kg feed. Straube *et al.* (1980) reported decrease (12-50%) in body weight in ferrets with dietary inclusion of ZnO @ 1500ppm and 3000ppm. Maita, *et al*. (1981) also reported decrease in body weight in rats at high doses of zinc sulphate @ 3000ppm and 30,000 ppm for a period of 13 weeks.

Table 1. Average body weight (gm) in different groups of experimental birds taken at 14 day interval (Mean ± SE)

Days of observation	C	G1	G2	G3
0 day	383 ± 13^{a}	363 ± 13.379^{a}	432 ± 48.02^{a}	393 ± 28.96^{a}
14	571 ± 27.49^{a}	529 ± 27.31^{a}	525 ± 49.54^{a}	402 ± 4.89^{b}
28	656 ± 12.08^{a}	591 ± 7.81^{b}	544 ± 4^{c}	416 ± 11.66^{d}
42	692 ± 3.74^{a}	640 ± 4.47^{b}	591 ± 10.53^{c}	420 ± 11.51^{d}
56	896 ± 6.78^{a}	854 ± 6.52^{b}	721 ± 14.17^{c}	480 ± 8.36^{d}

a,b,c,d; mean values in rows with no common superscripts differ significantly ($p<0.05$)

In the present study a relative decrease in weights of liver, spleen and pancreas was observed in all experimental groups (G1, G2 and G3) however, in group G3 a significant increase in relative organ weights of liver, spleen and pancreas was observed, however spleen does not show any variation in weight against group G2 (Table 2). The decrease in relative weight of different organs may be attributed to the proportionate decrease in body weights of experimental birds. Maita *et al.* (1981) also observed significant decrease in absolute and relative weights of liver, spleen, kidney, brain, heart and muscle in mice at 30,000 ppm zinc sulphate in feed for a period of 13 weeks. Hermayer *et al.* (1977) and Palafox *et al.* (1980) also demonstrated a marked reduction in body weight in poultry with dietary zinc @ 10 g and 20 g /kg zinc diet. Increase in the relative weight of different organs in group G3 at the end of experimental trail might be due to excessive loss of body weight in the experimental birds.

Table 2. Organ/body weight ratio percent (Mean± SE) of liver, pancreas and spleen in different groups of experimental birds

Organ	C	G1	G2	G3
Liver	2.76 ± 0.008^{w}	2.40 ± 0.14^{x}	$2.26\pm0.0.018^{y}$	2.31 ± 0.06^{z}
Pancreas	0.28 ± 0.005^{w}	0.20 ± 0.026^{x}	0.23 ± 0.008^{x}	0.26 ± 0.012^{w}
Spleen	0.14 ± 0.008^{w}	0.09 ± 0.005^{x}	0.11 ± 0.011^{x}	0.11 ± 0.008^{x}

w,x,y,z: mean values in rows with no common superscripts differ significantly($p<0.05$)

Macroscopically severe congestion in liver with marked atrophy of liver, spleen, bursa of fabricius, pancreas, gizzard and testes was highly appreciated in group G2 and G3. [Fig. 1(1-6)]

In Groups G1, G2 and G3, following microscopic lesions were observed. Liver showed degeneration of hepatocytes, distortion of hepatic cords and dilatation of hepatic sinusoids. Spleen revealed extensive haemorrhages. Koilin layer of gizzard showed tattered vertical rods with infiltrated heterophils. In bursa of fabricius cystic degenerative changes and depletion of lymphocytes in lymphoid follicles was noticed. In kidney extensive degeneration and necrosis of tubular epithelium and presence of solid dense coagulated mass of protoplasm in tubules was observed. Pancreas revealed Pancreas revealed dilatation of acinar lumina and dissorted acini. Testes did not revealed any microscopic change [Fig. 2(A-F)]

Similar lesions were also reported by Dewar *et al.* (1983) who found discolouration of koilin layer of gizzard, excessive desquamation of epithelial cells and lesions of exocrine pancreas in chicks with administration of high level of dietary Zn @ 2000, 4000 and 6000 ppm Zn/kg feed for duration of 4 weeks. Vanvleet *et al.* (1981) reported pancreatic necrosis in ducklings at high levels of zinc administration.

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REFERENCES

Creger, C.R. and Scott, J.T. 1977. Dietary zinc as an effective resting agent for the laying hen. *Poult. Sci*., **56**: 1706.

Dewar, W.A., Wight, P.P.I., Pearson, R.A. and Gentle, M.J. 1983. Toxic effects of high concentrations of zinc oxide in the diet of chicks and laying hens. *Br. Poult. Sci*., **24**: 397-404.

Gentle, M.J., Dewar, W.A., Wight, P.A.L. and Dick, K.M. 1982. The effects of dietary zinc on food intake in the domestic fowl. *appetite*: *J. of Intake Res.,* **3**: 53-60.

Hermayer, K.L., Stake, P.E. and Shippee, R. L. 1977. Evaluation of dietary zinc, cadmium, tin, lead, bismuth and arsenic toxicity in hens, *Poult. Sci.,* **56**: 1721-1722.

Johnson, D. Jr., Mehring, A.L., Savino, F.X. jr. and Titus, H.W. 1962. The tolerance of growing chickens for dietary. zinc. *Poult. Sci*., **41**: 311–317.

Kincaid, R.L., Miller, W.J., Jensen, L.S., Hampton, D.L., Neathery, M.W. and Gentry, R.P. 1976. Effect of high levels of dietary zinc and age upon tissue zinc in young chicks *Poult. Sci.,* **53**: 1954-1957.

Maita, K., Hirano, M., Mitsumori, K., Takahashi, K. and Shirasu, Y. 1981. Subacute toxicity studies with zinc sulfate in mice and rats. *J. Pestic. Sci.,* **6**: 327-336

National Research Council, 1994. Nutrient Requirements of Poultry. 9th rev.ed. National Academy Press, Washington, DC. 20418.

Oh, S.H., Nakave, H., Deagen, J.T., Whanger, P.D. and Arscott, G.H. 1979. Accumulation and depletion of zinc in chick tissue and metallothionins. *J. Nutr.,* **109**: 1720-1729.

Palafox, A.L., Ho-a, E. and Arch Tierz. 1980. Effect of zinc toxicity in laying white leghorn pullets and hens. *Poult. Sci.*, **59**: 2024-8.

Shippee, R.L., Stake, P.E., Koehn, N.V., Lambert, J.L. and Simmons, R.W. 1979. High dietary zinc or magnesium as forced resting agents for laying hens. *Poult. Sci.* **58**: 949-954.

Snedecor, G.W. and Cochran, W.G. 1989. Statistical Msethods. 8th Edition. Lowa state university press, lowa.

Straube, E.F., Schuster, N.H. and Inclair, A.J. 1980**.** Zinc toxicity in the ferret. *J. Comp. Pathol.,* **90**: 355-361.

Vanvleet, J.F., Boon, G.D. and Ferransv, I. 1981. Induction of lesions of selenium-vitamin E deficiency in ducklings fed silver, copper, cobalt, tellurium, cadmium or zinc: protection by selenium or vitamin E supplements. *Am. J. Vet. Res*., **42**: 1206-1217.