

Transport Stress and Alleviating its Effect by Supplemental Copper and Zinc in Laying Quails

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Abstract: This study was conducted to determine the effect of copper, zinc and their combinations on egg yield and reproductive performance of quails exposed to transport stress. In the study 80 day-old, 48 male and 144 female laying Japanese quails were randomly assigned to 1 control and 3 treatment groups, 3 replicates each consisting of 16 birds. The control group was fed a basal diet and the treatment groups were fed the basal diet supplemented with 150 mg of Cu kg⁻¹ of (group I), 150 mg of Zn kg⁻¹ of (group II), or 150 mg of Cu plus 150 mg of Zn kg⁻¹ (group III), during trial. The mean feed consumption ($p < 0.05$), final live weight ($p < 0.01$) and egg weight ($p < 0.01$) of the control group were lower than the treatment groups after transportation. In all groups transport stress caused a sudden decrease of hen day egg production, which reached to the normal level at the 2nd week in groups I and III, at the 3rd week in group II and at the 4th week in control group after transportation. Hatchability of fertile eggs were significantly higher at the 2nd ($p < 0.01$) and 3rd week ($p < 0.05$) after transportation in treatment groups. However, embryonic mortality was not significantly different between groups. The results suggested that transport and different environmental stress were ameliorated by supplementation of copper, zinc or their combination.

Key words: Transport stress, copper, zinc, quail, alleviating, supplemental

INTRODUCTION

Exposure of birds to stress is an inevitable event in poultry husbandry. Generally the term Stress is used to describe the detrimental effects of various factors on the health and performance of poultry. When the threshold level of stress is crossed it results in distress and birds became fatigued and weak. These conditions lead to birds' starvation and infectious disease (Dohms, 1990; Freeman, 1987).

Birds have limited body resources for growth, reproduction response to environmental changes and defense mechanism (Rosales, 1994). Domestic birds are subjected to frequent transport stress and therefore, it is important to have an effective management program to minimize their effects on the performance and health. Environmental stress has been shown to increase mineral excretion (El Husseiny and Creger, 1981; Smith and Teeter, 1987) while decreasing serum and tissue levels of antioxidant vitamins (Halliwell and Gutteridge, 1989; Sahin *et al.*, 2002; Sahin and Kucuk, 2003). Serum, liver and spleen levels of zinc are reduced in stressed birds

(Sahin *et al.*, 2002). Under the stress conditions, there is redistribution of body resources including energy and protein at the cost of decreased growth, reproduction and health (Brake, 1987). Dietary modifications are among the most preferred and practical ways to alleviate the effect of high environmental stress on poultry performance (Sahin and Kucuk, 2003).

Antioxidants minerals such as zinc have been used to ameliorate the effects of environmental stress (Lin *et al.*, 2002; Sahin, *et al.*, 2002; Sahin and Kucuk, 2003). Supplemental zinc is used in poultry diets because of its reported benefits to laying hens during periods of environmental stress (Onderci *et al.*, 2003; Sahin *et al.*, 2002; Smith and Teeter, 1987). Zinc has multiple important functions as a cofactor for more than 200 enzymes. Oxidative damage of the cell membrane by free radicals occurs during zinc deficiency (Prasad and Kucuk, 2002; Salgueri *et al.*, 2000). Furthermore, zinc can occupy iron and copper binding sites on lipids, proteins and DNA and thus exert a direct antioxidant action (Oteiza *et al.*, 1996; Tate *et al.*, 1999).

Biologically, copper is also an essential trace mineral for healthy animals and humans, yet it has a strong biological property. Zinc and Copper use has recently increased as a feed additive for promoting the growth of animals especially in the poultry (Jackson *et al.*, 1980; Sahin and Kucuk, 2003). Copper is routinely supplemented to swine and poultry diets at concentrations above the requirement of the animal, because pharmacological concentrations of Cu act as a growth stimulant in these species (Cromwell *et al.*, 1989; Lin *et al.*, 2002). Low intakes of several minerals including copper and zinc have also been implicated with abnormal lipid profiles (Bakalli *et al.*, 1995; Sahin and Kucuk, 2003; Smith and Teeter, 1987). Many studies have reported that zinc induced copper deficiency (Summerfield *et al.*, 1992) as well as copper induced zinc deficiency (Hermann *et al.*, 1998; Mikhail *et al.*, 1996; Sandstead, 1995). In a study to determine the tolerable level of copper in laying hen diets, Jackson (1977) found that total feed intake, water consumption and egg production were not negatively influenced by 480 mg kg⁻¹ of dietary copper. Egg production, however, decreased to half that of the control at a level of 960 mg kg⁻¹ and ceased after feeding 1920 mg kg⁻¹ of dietary copper for 14 days. In a 336 day feeding trial, Jackson *et al.* (1979) added various levels of copper to a practical diet. These authors reported a decrease in body weight gain, egg production and various tissue weights at a level of 600 mgkg⁻¹. They also reported no adverse effect on the reproductive system at a level of 800 mgkg⁻¹. However, feather growth and egg production decreased to half that with the control ration.

Transport stress is a physical stress (Jones *et al.*, 1988; Gregory *et al.*, 1992) and regarding the effect of transport stress on laying quails is very limited.

MATERIALS AND METHODS

According to their initial body weights 80 day old 48 male and 144 female Japanese quails were randomly assigned to 1 control and 3 treatment groups, 3 replicates

each consisting of 16 birds (4 male and 12 female). The birds were fed either a basal diet or the basal diet supplemented with 150 mg of Cu kg⁻¹ (group I), 150 mg of Zn kg⁻¹ (group II) and 150 mg of Cu plus 150 mg of Zn kg⁻¹ (group III). The mineral mixes were prepared from reagent grade or ultra pure chemicals. Zinc sulfate and copper sulfate were used as mineral sources. Chemical compositions of the diets were analyzed using the international procedures of AOAC (1984). Ingredients and chemical composition of the basal diet are shown in Table 1. A small part of the basal diet were first mixed with the respective amounts of Copper, Zinc and their combinations as a small batch and then with a larger amount of the basal diet until the total amount of the respective diets were homogeneously mixed.

Light regime was 16 h/day. The diets and fresh water were offered ad libitum. After ten days of beginning to trial, all groups have been transferred to a different place in carton boxes by car about 25 km away from their previous place. Temperature and humidity in the poultry house was recorded 12 times a day with a TESTO 175 electronic instrument. Feed consumptions were recorded at weekly intervals for each group and feed conversion ratio was not calculated because of both sex were in the same cage.

The eggs of all groups were collected daily and weighed throughout the experiment. One third of collected eggs in each group stored at 15°C for 1 day and incubated at 37, 5°C; 70% RH. After hatching; all chicks were weighed and the number of chicks and unhatched eggs were recorded. Then the numbers of fertile, infertile eggs, total death embryos were determined by breaking of the unhatched eggs. Body weights of laying quails were recorded at the beginning and at the end of the study.

All proportional data were transformed to arcsine before analysis. Data of live weights, chick weights, egg weights, daily egg production, hatchability and total dead embryos, were statistically analyzed by One-Way ANOVA test between groups and Two-Way ANOVA test between weeks. Differences among treatments were separated by Duncan's test in SPSS (1998).

Table 1: Ingredients and chemical composition of the basal diets

Ingredients	(gkg ⁻¹)	Chemical composition	(gkg ⁻¹)
Yellow Corn	465	Dry matter	895.8
Wheat	85	Crude protein	207.0
Soybean meal	293	Calcium	33.8
Fish meal	15	Total phosphorus	5.6
Vegetable oil	46.0	Calculated values	
Calcium Carbonate	77	ME (MJ kg ⁻¹)	12.53
Dicalcium Phosphate	13	Lysine	11.2
Salt	3	Methionine+Cystine	7.1
DL-methionine	0.5		
Vitamin mineral premix ^a	2.5		

^aVitamin mineral premix (provided the following per kg diet): Vitamin A, 12500 IU; Vitamin D3, 1500 IU; Vitamin E, 31.25 mg; Vitamin K3, 3.75 mg; Vitamin B1, 2.5 mg; Vitamin B2, 7.5 mg; Niacin 25 mg; Cal. D-pantothenate 10 mg; Vitamin B6, 5mg; Vitamin B12, 0.019 mg; Folic acid 1 mg; Choline chloride 250 mg; Mn 100 mg; Fe 75 mg; Zn 75 mg; Cu 6.25 mg; Co 0.25 mg; I, 1.25 mg; Se 0.19mg

RESULTS AND DISCUSSION

Table 2 presents the effect of Cu, Zn, and Cu+Zn supplementation to diet on live weight, egg weight and feed consumption in laying quails before and after transport. Experimental results indicated that final live weight ($p<0.01$), feed consumption throughout the trial ($p<0.05$) and egg weight after ($p<0.01$) transport were positively affected by Cu, Zn and Cu+Zn supplementation. Initial live weight and egg weights of the birds before transportation were not affected by dietary Cu, Zn or Cu+Zn supplementation of laying quails. Average ambient relative humidity and mean value of daily temperature in the hen house before and after transportation were 46 ± 5.6 and 48 ± 4.1 %, 20 ± 3.6 and 21 ± 2 , 8°C , respectively.

The data on hen day egg production before and after transport in laying quail are shown in Table 3. Hen day egg productions in treatment groups were significantly higher than the control group during three weeks after transportation. Transport and different environmental

stress caused a sudden fall in the egg yield in all groups compared with the egg yield before transportation. Hen day egg production of group I and group III reached to the normal level at 2nd week, group II at 3rd week and control group at 4th week after transportation. There were significant differences between weeks of all groups. The results showed that Cu supplementation is more effective than Zn in transport stress.

Transport stress decreased hatchability of fertile eggs which was elevated by supplemental Cu, Zn and Cu+Zn (Table 4). Hatchability of fertile eggs was significantly higher at the 2nd and 3rd week after transportation in trial groups. The results of the current study indicated that supplementation of diets with Cu, Zn and Cu+Zn improved 2nd and 3rd week's hatchability of fertile eggs ratio without affecting embryonic death in laying Japanese quail ($p<0.01$ and $p<0.05$).

There are very limited documents on beneficial effect of Cu and Zn supplementation to the diets of laying Japanese quails exposed to transport stress. Sahin and Kucuk (2003) reported that adding 30 and 60 mg kg^{-1} of

Table2: Effect of Copper, Zinc and their combinations on live weight, egg weight and feed consumption

Item	Cu 150 mgkg^{-1}	Zn 150 mgkg^{-1}	Zn 150 mgkg^{-1} + Cu 150 mgkg^{-1}	Control	Pooled SEM	P-value
Initial live weight (g)	173.32	174.22	175.44	175.50	0.65	$p>0.05$
Final live weight (g)	176.52 ^a	175.03 ^a	175.89 ^a	169.50 ^b	0.67	$p<0.01$
Egg weight before transportation (g)	10.26	10.15	10.20	10.16	0.02	$p>0.05$
Egg weight after transportation (g)	10.26 ^a	10.14 ^{ab}	10.22 ^a	10.10 ^b	0.01	$p<0.01$
Mean feed consumption throughout the trial (g)	26.05 ^a	26.32 ^a	25.90 ^a	24.50 ^b	0.25	$p<0.05$

a-e within rows, means followed by different letters are significantly different

Table 3: Effect of Copper, Zinc and their combinations on Egg production

Egg production, hen day (%)	Cu 150 mgkg^{-1}	Zn 150 mgkg^{-1}	Zn 150 mgkg^{-1} + Cu 150 mgkg^{-1}	Control	Pooled SEM	P-value
Before transport	81.8 _m	80.5 _m	80.5 _m	79.2 _m	1.62	$p>0.05$
1st week after transport	65.3 ^a _n	50.0 ^b _o	63.9 ^a _n	34.7 ^c _o	3.09	$p<0.001$
2nd week after transport	74.6 ^a _{mn}	63.5 ^{ab} _{no}	73.0 ^a _{mn}	41.2 ^b _o	3.27	$p<0.001$
3rd week after transport	79.2 ^a _m	76.4 ^a _{mn}	77.8 ^a _m	54.2 ^b _n	2.41	$p<0.001$
4th week after transport	80.6 _m	79.2 _{mn}	79.2 _m	70.87 _m	1.64	$p>0.05$
Pooled SEM	1.97	2.67	1.87	2.94		
P-value	$p<0.05$	$p<0.001$	$p<0.05$	$p<0.001$		

a-e within rows, means followed by different letters are significantly different .m-o within columns, means followed by different letters are significantly different

Table-4 Effects of Copper, Zinc and their combinations on Hatchability of eggs and embryonic death in laying quail before and after exposed to transport stress

Item	Cu 150 mgkg^{-1}	Zn 150 mgkg^{-1}	Zn 150 mgkg^{-1} + Cu 150 mgkg^{-1}	Control	Pooled SEM	P-value
Hatchability of eggs (%)						
Before transport	72.85	72.53	70.36	71.94 _m	0.82	$p>0.05$
1st week after transport	70.95 ^a	72.51	73.69	71.69 _m	0.79	$p>0.05$
2nd week after transport	73.13 ^a	69.21 ^a	68.47 ^a	64.07 ^b _o	0.66	$p<0.01$
3rd week after transport	73.52	72.50 ^a	71.41 ^a	66.62 ^b _{no}	0.84	$p<0.05$
4th week after transport	73.65	72.58	70.86	68.60 _{mn}	0.72	$p<0.05$
Pooled SEM	0.72	0.67	0.66	0.70		
P-value	$p>0.05$	$p>0.05$	$p>0.05$	$p<0.01$		
Total death embryos (%)						
Before transport	25.47	23.90	22.50	24.14	0.54	$p>0.05$
1st week after transport	23.68	22.60	25.32	23.50	0.53	$p>0.05$
2nd week after transport	25.52	23.63	25.40	26.05	0.50	$p>0.05$
3rd week after transport	24.52	23.91	24.68	23.50	0.48	$p>0.05$
4th week after transport	23.34	21.91	22.23	22.87	0.56	$p>0.05$
Pooled SEM	0.50	0.43	0.47	0.49		
P-value	$p>0.05$	$p>0.05$	$p>0.05$	$p>0.05$		

a-e within rows, means followed by different letters are significantly different. m-o within columns, means followed by different letters are significantly different

CuSO₄H₂O to diets of laying quails rearing under high temperature stress resulted in a higher egg production and egg quality compared to control groups. Ýpek *et al.* (2003) reported that Zn and Cu fed at 150 ppm level enhanced egg production and egg weight of laying Japanese quail. Sahin *et al.* (2002) reported that Zn supplementation improved egg production and quality in laying hens reared under low ambient temperature. Avçi *et al.* (2003) reported that diet supplemented with Zn improved body weight gain and feed efficiency without affecting feed consumption and blood parameters in Japanese quails. The results of this study are in agreement with the other data obtained from studies on laying Japanese quails (Jones *et al.*, 1988; Prasad and Kucuk, 2002; Tate *et al.*, 1999) and laying hens Gregory *et al.* (1992). Environmental stress increases free radical production and lowers the concentration of antioxidant minerals such as Zn and Cr in serum (Halliwell and Gutteridge, 1989). Beisel (1982), Sahin *et al.* (2002) and Shaheen and Abd El-Fattah (1995) reported that dietary Zn deficiency caused an increased lipid peroxidation and this was inhibited by Zn supplementation in rats. Hermann *et al.* (1997) reported that dietary Cu and Zn have significant interactive effect on plasma triglyceride concentration. Moreover, stress causes an accumulation of Zn in the liver, decreasing plasma zinc concentration; thus it may exacerbate a marginal zinc deficiency or an increased Zn requirement. Heat stress also causes an increase in the excretion of minerals such as zinc, copper and manganese (Beisel, 1982). In the present study transport stress significantly depressed feed intake. Therefore, the decrease of egg production, egg weight and hatchability might be resulted from the decrease of feed intake due to transport stress. The reduction of feed consumption and increase in Cu and Zn excretion under environmental stress adversely affects poultry performance, health status and antioxidant system (Sahin and Kucuk, 2003; Sahin *et al.*, 2002).

CONCLUSION

The results of the current study suggested that supplementation of diets with Cu and Zn separately or as a combination offers an alternative way to reduce the loss of performance in laying Japanese quails exposed to transport stress.

REFERENCES

AOAC, 1984. Official Methods of Analysis of the Association of Official Analytical Chemistry. 14th Edn. Virginia.

- Avçi, M., M. Yerturk and H. Ýpek, 2003. The affect of Zn supplementation on growth performance and some hematological parameters in quail diets. *J. Faculty of Veterinary medicine. University of Yuzuncu Yýl.*, 1: 61-64.
- Bakalli, R.I., G.M. Pesti, W.L. Ragland and V. Konjufca, 1995. Dietary copper in excess of nutritional requirement reduces plasma and breast muscle cholesterol of chickens. *Poult. Sci.*, 74: 360-365.
- Beisel, W.R., 1982. Single nutrients and immunity. *Am. J. Clin. Nutr.*, 35: 442-451.
- Belay, T. and R.G. Teeter, 1987. Effects of environmental temperature on broiler mineral balance partitioned into urinary and fecal loss. *Br. Poult. Sci.*, 37: 423-433.
- Brake, J.T., 1987. Stress and Modern poultry management. *Annuals production highlights. F. Hoffman-La Roche and Co. Ltd. 4002. Basie, Switzerland.*
- Cromwell, G.L., T.S. Stahly and H.J. Monegue, 1989. Effects of source and level of copper on performance, and liver copper stores in weanling pigs. *J. Anim. Sci.*, 67: 2996-3002.
- Dohms, J.E., 1990. Stress Mechanisms of immune suppressions. In: *Proc. 41st North Central Avian Dis. and Poultry immunosuppressive Dis. Symp.*, Columbus. OH, pp: 22-42.
- El Husseiny, O. and C.R. Creger, 1981. Effect of ambient temperature on mineral retention and balance of the broiler chicks. *Poult. Sci.*, 60: 1651 (Abst.).
- Freeman, B.M., 1987. The Stress Syndrome. *World's Poult. Sci. J.*, 43: 15-19.
- Gregory, N.G., L.J. Wilkins and S.D. Austin *et al.*, 1992. Effect of catching method on the prevalence of broken bones in end of lay hens. *Avian Pathology*, 21: 717-722.
- Halliwell, B.E. and J.M.C. Gutteridge, 1989. Lipid peroxidation: A radical chain reaction. In *Free Radicals in biology and Medicine*. 2nd Edn. 188-218 Oxford University Press. New York.
- Hermann, J., C. Goad, A. Arquit, B. Stoecker, R. Porter, H. Chung and P.L. Claypool, 1998. Effects of dietary Chromium, Copper and Zinc on plasma lipid concentrations in male Japanese quail. *Nutr. Res.*, 18: 1017-1027.
- Hermann, J., C. Goad, B. Stoecker, A. Arquitt, R. Porter, B. Adeleye P.L., Claypool and G. Brusewitz, 1997. Effects of dietary chromium, copper and zinc on femur fracture force and femur calcium concentration in male Japanese quail. *Nutr. Res.*, 12: 1529-1540.
- Jackson, N., M.H. Stevenson and G.M. Kirkpatrick, 1979. Effects of the protracted feeding of copper sulfate-supplemented diets to laying domestic fowl on egg production and on specific tissues, with special reference to mineral content. *Br. J. Nutr.*, 42: 253-266.

- Jackson, N., 1977. The effect of dietary copper sulfate on laying performance, nutrient intake and tissue copper and iron levels of the mature, laying domestic fowl. *Br. J. Nutr.*, 38: 93-100.
- Jackson, N., M.H. Stevenson and G.M. Kirkpatrick, 1980. Effects of level of dietary copper sulphate and period of feeding on the laying domestic fowl, with special reference to tissue mineral content. *Br. J. Nutr.*, 43: 205-215.
- Jones, R.B., G. Beuving and H.J. Blokhuis, 1988. Tonic Immobility and Heterophil/ Lymphocyte Responses of the Domestic Fowl to Corticosterone Infusion. *Physiol. Behav.*, 42: 249-253.
- Lin, H., L.F. Wang, J.J. Song, Y.M. Xie and Q.M. Yang, 2002. Effect of dietary supplemental levels of vitamin A on egg production and immune responses of heat-stressed laying hens. *Polt. Sci.*, 81: 458-465.
- Mikhail, T.H., W.G. Nicola K.H. Ibrahim, S.H. Saiama and M. Emam, 1996. Abnormal zinc and copper metabolism in hepatic steatosis. *Boll. Chim. Farm.*, 135: 591-597.
- Onderci, M., N. Sahin, K.Sahin and N. Kilic, 2003. The antioxidant properties of chromium and zinc: *In vivo* effects on digestibility, lipid peroxidation, antioxidant vitamins and some minerals under a low ambient temperature. *Biol. Trace Elem. Res.*, 92: 139-150.
- Oteiza, P.I., K.L. Olin., C.G. Fraga and C.L. Keen, 1996. Oxidant defense systems in testis from Zn deficient rats. *Proc. Soc. Exp. Biol. Med.*, 213: 85-91.
- Prasad, A.S. and O. Kucuk, 2002. Zinc in cancer prevention. *Cancer metastasis Rev.*, 21: 291-295.
- Rosales, A.G., 1994. Managing Stress in Broiler. *J. Applied Poult. Res.*, 3: 199-207.
- Sahin, K. and O. Kucuk, 2003. Zinc supplementation alleviates heat stress in laying Japanese quail. *J. Nutr.*, 133, 9; Health Module, pp: 2808.
- Sahin, K., O. Kucuk, N. Sahin and M. Sari, 2002. Effect of vitamin C and vitamin E on lipid peroxidation status, some serum hormone, metabolite and mineral concentrations of Japanese quails reared under heat stress. *Ynt. J. Vitam. Nutr. Res.*, 72: 91-100.
- Sahin, N., M. Onderci and K. Sahin, 2002. Effects of dietary chromium and Zinc on egg production, egg quality and some blood metabolites of laying hens reared under low ambient temperature. *Biol. Trace Elem. Res.*, 85: 47-58.
- Salgueri, M.J., M. Zubillaga, A. Lysionek, M.I Sarabia, R. Caro, T. De Paoli, A.Hager, R. Well and J. Boccio, 2000. Zinc as essential micronutrient. *A Rev. Nutr. Res.*, 20: 737-755.
- Sandstead, H.H., 1995. Requirements and toxicity of essential trace elements, illustrated by zinc and copper. *Am. J. Clin. Nutr.*, 61: 621-624.
- Shaheen, A.A. and A.A. Abd El-Fattah, 1995. Effect of dietary zinc on lipid peroxidation, glutathione, protein thiols levels and superoxide dismutase activity in rat tissues. *Int. J. Biochem. Cell. Biol.*, 27: 89-95.
- Smith, M.O. and R.G. Teeter, 1987. Potassium balance of the 5-8 week old broiler exposed to constant heat or cycling high temperature stress and the effects of supplemental potassium chloride on body weight gain and feed efficiency. *Poult. Sci.*, 66: 487-492.
- SPSS., 1998. Spss for Windows 9.3 Base System User's Guide. Release 9.0 Copyright by SPSS Inc. Printed, USA.
- Summerfield, A.L, F.U. Steinberg and J.G. Gonzalez, 1992. Morphologic findings in bone marrow precursor cells in zinc-induced copper deficiency anemia. *Am. J. Clin. Pathol.*, 97: 665-668.
- Tate, D.J., M.V. Miceli and D.A. Newsome, 1999. Zn protects against oxidative damage in cultured human retinal pigment epithelial cells. *Free Radic. Biol. Med.*, 26: 704-713.
- İpek, H., M. Yerturk and M. Avçi, 2003. The effect of supplemental Zn and Cu on some egg yield characters and hematological parameters in laying quails. *Journal of Faculty of Veterinary medicine. Univ. of Yuzuncu Yıl.*, 1: 65-68.