

Influence of dietary supplementation of selenium and vitamin E on bone morphometry and mineralization in broiler chicken under heat stress

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Abstract

A study on bone development under the influence of selenium and vitamin E supplementation was under taken in broiler chicken. Three levels of each selenium (0.15, 0.35, and 0.45 mg/kg diet) and vitamin E (50, 100, and 150 IU/ kg diet) were taken to formulate nine treatment groups in the form of broiler starter and finisher to evaluate the bone morphometry and bone mineralization of broiler chicken. The experiment was conducted in a high temperature humidity index (83.94) during the August-September period. The results revealed that bone morphometry parameters such as tibia bone proximal width, mid shaft width, distal width, and length were not influenced by dietary selenium and vitamin E supplementation. Similarly, the mean tibia bone weight (g), bone moisture (%), bone calcium (%), bone phosphorus (%), total ash (%), and bone Zn content (mg/kg) did not differ significantly due to selenium and vitamin E levels in the diet. Therefore, it can be concluded that the bone morphometry and bone mineralization in broiler chicken is not affected by different levels of selenium and vitamin E in the diet.

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1. Introduction

The exposure to thermal stress in poultry causes uncontrolled free radicals and reactive oxygen species generation which result in oxidative damage and physiological disturbances in cellular functions in the body (Zhang et al. 2014; Emami et al. 2020). Along with the compromised growth performance thermal stress causes excessive fat deposition and reduced bone mineralization in broiler chicken (Hosseini-Vashan et al. 2016; Emami et al. 2021). Drastic reduction in availability of calcium and phosphorus has been reported in broiler chicken subjected to heat stress which negatively affects bone development (Sgavioli et al. 2016). For economic viability of poultry farming and bird welfare bone health is an important issue (Shim et al. 2012). Under heat stress, as a mechanism of heat dissipation, the increased respiratory rate causes respiratory alkalosis and high blood pH which reduces blood calcium level and in turn results in lower bone mineralization

and bone strength. Furthermore, the exposure to heat stress results in elevated corticosterone which enhances the osteoclastic activity in the bones and lowers the mineral absorption in the body (Xu et al. 2018; Yan et al. 2019).

Just like other animals, birds execute different internal defense mechanisms to overcome the negative effects of the heat stress. And, such internal defense mechanisms of the birds against the heat stress can be complemented by external environmental and nutritional interventions. The nutritional interventions include supplementation of vitamins, minerals, and antioxidants (Habibian et al. 2016; He et al. 2018). The antioxidant properties of selenium have improved the growth performance and physiological parameters of broiler chicken by reducing the effects of oxidative stress imposed by high environmental temperature (Choct et al. 2010; Habibian et al. 2014). Selenium was shown to improve the biomechanical strength of bones with higher modulus of elasticity. On similar

lines, vitamin E has shown significant antioxidant properties which scavenges free radicals and prevents the cells from oxidative damage (Traber and Atkinson 2007; Gao et al. 2010; Surai et al. 2019). On vitamin E supplementation improved growth performance and reduced oxidative stress have been reported in broiler chicken reared under high temperature conditions (Sahin et al. 2002; Attia et al. 2017). Vitamin E supplementation increases the rate of bone formation by protecting the osteoblasts from the oxidative damage of free radicals (Xu et al. 1995). However, excess vitamin E level exerts negative effect on the vitamin D3 utilisation (Aburto and Britton 1998). Therefore, to understand the synergistic effect of selenium and vitamin E this study was conducted to evaluate the effects of selenium and vitamin E supplementation on the bone morphometry and mineralization in broiler chicken.

2. Materials and methods

A experiment was conducted in a (3x3) factorial design in broiler chicken with three levels of selenium (0.15, 0.30, and 0.45 mg/kg diet) and three levels of vitamin E (50, 100, and 150 IU/kg diet) resulting in nine dietary treatments. A total of 288 day-old chicks of uniform body weight were used for the experiment and each treatment was allocated four replicates of chicks with eight chicks in each replicate. The birds were reared in battery cages for a period of six weeks in which birds had free access to clean drinking water and feed. The birds were provided with starter and finisher diets during 0-3 weeks and 4-6 weeks of age, respectively (Table 1). The experiment was conducted under the conditions of relatively higher temperature humidity index (83.94 ± 0.36) during the August-September period.

At the end of the feeding trial eight birds from each treatment group (2 bird/ replicate) were taken at random and slaughtered after 12 hours of fasting with *ad lib* drinking water for harvesting the tibia bones of the carcasses. The associated connective tissue was thoroughly and carefully separated from the bones and all the bones were properly labelled for identification of treatment group. The bones were dipped in boiling water for five minutes for removing the remaining pin and soft tissue. The morphometry on the bone were carried according to Collins and Moran (1999). The length, mid-shaft width, proximal epiphysis and distal epiphysis width of the tibia bone were measured with vernier calipers. Same bones were further used for the study of bone mineralization. Each tibia bone was defatted by dipping in petroleum spirit (boiling point 60-80 °C) for 16 hours using the Soxhlet apparatus followed by drying. The bone weight (fresh and dried) was taken followed by ashing in muffle furnace for estimation of bone ash, bone calcium, bone phosphorus, and bone zinc. The data pertaining to bone morphology and mineralization were analyzed by two way ANOVA using the General linear model of SPSS-20.

3. Results

Table 1 Broiler starter and finisher diet ingredients and nutrient composition

Ingredients (%)	Starter (0-21 days)	Finisher (22-42 days)
Maize	53.57	61.65
Soya bean meal	42.0	34.0
Oil	1.0	1.3
Lime stone	0.92	1.1
Di-calcium phosphate	1.73	1.285
Salt	0.30	0.3
DL-Methionine	0.165	0.05
TM-Premix ¹	0.10	0.10
Vit-Premix ²	0.15	0.15
B complex	0.015	0.015
Choline chloride	0.05	0.05
Total	100	100
Analyzed values		
Crude protein (%)	23.22	20.40
Calcium (%)	1.02	0.95
Total P (%)	0.72	0.66
Selenium (mg/kg)*	0.15	0.13
Calculated values		
Metabolizable energy (kcal/kg)	2905	3006
Available phosphorus (%)	0.45	0.36
Lysine (%)	1.25	1.06
Methionine (%)	0.53	0.38
Threonine (%)	0.99	0.87
¹ Trace mineral premix supplied mg/kg diet: Mg300, Mn 60, I0.4, Fe 80, Cu 8, Zn 40, Se*-variable		
² Vitamin mixture provided mg/kg diet: Choline chloride 500, Niacin12, Pyridoxine hydrochloride 1.6, vitamin A 82500IU, Vitamin D ₃ 2000IU, Vitamin B ₁ 0.8, Vitamin B ₂ 6, Vitamin B ₁₂ 8, Vitamin K 1, Vitamin E*-variable		
*Values variable in test diet		

3.1 Bone morphometry

The mean values of different bone morphometry traits influenced by feeding different levels of selenium and vitamin E are given in Table 2. No significant dietary effects were observed on tibia bone proximal width, mid shaft width, distal width, and length.

3.2 Bone Mineralization

The effect of different selenium and vitamin E levels on tibia bone mineralization is given Table 3. No significant dietary effects were observe don the tibia bone weight, moisture, ash, calcium, phosphorus, and Zn contents.

Table 2 Bone morphometry of broiler chicken under the influence of selenium and vitamin E supplementation

Treatment		Tibia bone morphometry			
Selenium (mg/kg)	Vitamin E (IU/kg)	Proximal width (mm)	Mid shaft width (mm)	Distal width (mm)	Length (mm)
0.15	50	10.05	7.51	7.96	90.63
	100	10.09	8.03	7.30	94.18
	150	9.99	7.51	7.13	95.43
0.30	50	10.11	7.61	7.95	94.09
	100	10.11	7.78	7.90	95.05
	150	9.88	7.59	7.55	93.73
0.45	50	10.13	7.73	7.80	95.15
	100	10.55	7.14	7.46	93.49
	150	10.28	7.21	7.70	92.99
Pooled SEM		0.109	0.080	0.069	0.426
Probability					
Selenium		NS	NS	NS	NS
Vitamin E		NS	NS	NS	NS
Interaction		NS	NS	NS	NS

NS: Non-significant

4. Discussion

Thermal stress is a challenging environmental factor in poultry production, particularly in tropical areas, with serious economic implications, such as growth performance, egg production, health of birds, metabolism of birds, and behavioural activities of birds. The skeletal disorders, tibial dyschondroplasia, under heat stress is another major problem in poultry industry which results in excess condemnation of poultry carcasses (Świątkiewicz and Arczewska-Włosek 2012). Imbalance of thyroid hormones have been implicated in the

pathogenesis of tibial dyschondroplasia in chicken under heat or oxidative stress. The reduction in the plasma levels of T3 and corresponding increase of T4 levels have been reported in broiler chicken under high temperature conditions and oxidative stress which negatively affect the bone tissue metabolism and the consequent bone strength (Williams 2009; Lara and Rostagno 2013; Boostani et al. 2015). Another negative aspect of heat stress is the dysregulation of calcium and phosphorus metabolism which are major determinants of bone mineralization and bone strength. Heat stress have been

Table 3 Bone mineralization of broiler chicken under the influence of selenium and vitamin E supplementation

Treatment		Tibia bone morphometry					
Selenium (mg/kg)	Vitamin E (IU/kg)	Fresh bone weight (g)	Moisture (%)	Calcium (%)	Phosphorus (%)	Total ash (%)	Zinc (ppm)
0.15	50	50	4.54	49.31	22.32	7.80	40.80
	100	100	4.43	51.10	24.35	7.43	38.35
	150	150	4.56	50.10	22.33	7.24	39.77
0.30	50	50	4.51	49.66	23.87	7.94	38.76
	100	100	4.47	50.64	24.29	6.71	42.57
	150	150	4.46	51.08	24.62	7.51	41.65
0.45	50	50	4.34	50.17	24.26	7.35	40.22
	100	100	4.58	50.69	24.14	7.56	38.66
	150	150	4.60	50.44	25.20	8.22	39.54
Pooled SEM		0.042	0.393	0.298	0.218	0.972	1.006
Probability							
Selenium		NS	NS	NS	NS	NS	NS
Vitamin E		NS	NS	NS	NS	NS	NS
Interaction		NS	NS	NS	NS	NS	NS

NS: Non-significant

shown to negatively affect the calcium and phosphorus levels in bones which hasten the development of tibial dyschondroplasia in chicken (Hosseini-Vashan et al. 2016).

However, in this study none of the bone morphometry and bone mineralization parameters were affected by supplementation of different levels of selenium and vitamin E in the diet of broiler chicken. On similar lines, the selenium supplementation was reported to have no influence on the calcium, phosphorus, and ash content of tibia bones (Ndubuisi et al. 2021), however, on contrary these authors reported increased bone weight and length in broiler chicken. Similarly, selenium supplementation had no effect on the tibia bone weight but resulted in higher bone calcium percentage which indicates improved bone mineralization in response to selenium supplementation (Attia et al. 2010). The selenium supplementation improved calcium deposition in tibia of layer hens (Attia et al. 2010) and enhanced cartilage integrity (Medeiros 2016). The selenium deficiency has been reported to cause growth retardation and modified bone metabolism which negatively affects the bone health (Zeng et al. 2013). With respect to vitamin E supplementation, the heat stress induced reduction of bone mineral content and bone mineral density was reversed by vitamin E/selenium supplementation (Calik et al. 2022). The vitamin E has been found protecting the bone forming cells, osteoblasts, from oxidative damage of free radicals which results in accretion of new bone mass by these cells (Xu et al. 1995). However, it is noteworthy to mention that the differences in results with respect to earlier studies may be because of differences in experimental conditions or because the levels of selenium as well as vitamin E in the basal diet are sufficient for development bone.

5. Conclusions

In this study it was concluded that the bone morphometry and bone mineralization in broiler chicken is not affected by different levels of selenium and vitamin E in the diet which may be because of the fact that the levels present in the basal diet are sufficient to support the normal bone development in broiler chicken.

Declarations

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References

Aburto A, Edwards HM Jr, Britton WM. (1998). The influence of vitamin A on the utilization and amelioration of toxicity of cholecalciferol, 25-hydroxycholecalciferol, and 1,25 dihydroxycholecalciferol in young broiler chickens. *Poultry Science* 77(4): 585-593.

- Attia YA, Abdalah AA, Zeweil HS, Bovera F, El-Din AT, Arafa MA. (2010). Effect of inorganic or organic selenium supplementation on productive performance, egg quality and some physiological traits of dual-purpose breeding hens. *Czech Journal of Animal Science* 55: 505-519.
- Attia YA, Al-Harhi MA, El-Shafey AS, Rehab YA, Kim WK. (2017). Enhancing tolerance of broiler chickens to heat stress by supplementation with vitamin E, vitamin C and/or pro-biotics. *Annals of Animal Science* 17: 1155-1169.
- Boostani A, Sadeghi AA, Mousavi SN, Chamani M, Kashan N. (2015). The effects of organic, inorganic, and nano-selenium on blood attributes in broiler chickens exposed to oxidative stress. *Acta Scientiae Veterinariae* 43: 1-6.
- Calik A, Emami NK, White MB, Walsh MC, Romero LF, Dalloul RA. (2022). Influence of dietary vitamin E and selenium supplementation on broilers subjected to heat stress, Part I: Growth performance, body composition and intestinal nutrient transporters. *Poultry Science* 101: 101857. <https://doi.org/10.1016/j.psj.2022.101857>
- Collins NE, Moran ET Jr. (1999). Influence of supplemental manganese and zinc on live performance and carcass quality of broilers. *Journal Applied Poultry Research* 8: 222-227.
- Emami NK, Greene ES, Kogut MH, Dridi S. (2021). Heat stress and feed restriction distinctly affect performance, carcass and meat yield, intestinal integrity, and inflammatory (chemo) cytokines in broiler chickens. *Frontiers in Physiology* 12: 707757. <https://doi.org/10.3389/fphys.2021.707757>
- Emami NK, Jung U, Voy B, Dridi S. (2020). Radical response: effects of heat stress-induced oxidative stress on lipid metabolism in the avian liver. *Antioxidants* 10(1): 35. <https://doi.org/10.3390/antiox10010035>
- Gao J, Lin H, Wang XJ, Song ZG, Jiao HC. (2010). Vitamin E supplementation alleviates the oxidative stress induced by dexamethasone treatment and improves meat quality in broiler chickens. *Poultry Science* 89: 318-327.
- Habibian M, Ghazi S, Moeini M. (2016). Effects of dietary selenium and vitamin E on growth performance, meat yield, and selenium content and lipid oxidation of breast meat of broilers reared under heat stress. *Biological Trace Element Research* 169: 142-152.
- He S, Arowolo M, Medrano R, Li S, Yu Q, Chen J, He J. (2018). Impact of heat stress and nutritional interventions on poultry production. *World Poultry Science Journal* 74: 647-664.
- Hosseini-Vashan S, Golian A, Yaghoobfar A. (2016). Growth, immune, antioxidant, and bone responses of heat stress-exposed broilers fed diets supplemented with tomato pomace. *International Journal of Biometeorology* 60: 1183-1192.
- Lara LJ, Rostagno MH. (2013). Impact of heat stress on poultry production. *Animals* 3: 356-369.
- Medeiros DM. (2016). Copper, iron, and selenium dietary deficiencies negatively impact skeletal integrity: a review. *Experimental Biology and Medicine* 241: 1316-1322.
- Ndubuisi DI, Daudu OM, Abdurashid M. (2021). The role of selenium in ameliorating the impact of heat stress on growth, hormones, minerals and tibia measurements of broiler chickens. *Nigerian Journal of Animal Production* 48(4): 176 -184.
- Sahin K, Kucuk O, Sahin N, Gursu M. (2002). Optimal dietary concentration of vitamin E for alleviating the effect of heat stress

- on performance, thyroid status, ACTH and some serum metabolite and mineral concentrations in broilers. *Veterinary Medicine-Czech* 47: 110-116.
- Sgavioli S, Domingues CHF, Santos ET, Quadros TCO, deBorges LL, Garcia RG, Louzada MJ QL, Boleli IC. (2016). Effect of in-ovo ascorbic acid injection on the bone development of broiler chickens submitted to heat stress during incubation and rearing. *Revista Brasileira de Ciencia Avicola* 18(1): 153-162.
- Shim MY, Karnuah AB, Mitchell AD, Anthony NB, Pesti GM, Aggrey SE. (2012). The effects of growth rate on leg morphology and tibia breaking strength, mineral density, mineral content, and bone ash in broilers. *Poultry Science* 91: 1790-1795.
- Surai PF, Kochish II, Romanov MN, Griffin DK. (2019). Nutritional modulation of the antioxidant capacities in poultry: the case of vitamin E. *Poultry Science* 98: 4030-4041.
- Swiatkiewicz S, Arczewska-Wlosek A. (2012). Bone quality characteristics and performance in broiler chickens fed diets supplemented with organic acids. *Czech Journal of Animal Science* 57: 193-205.
- Traber MG, Atkinson J. (2007). Vitamin E, antioxidant and nothing more. *Free Radical Biology and Medicine* 43: 4-15.
- Williams GR. (2009). Actions of thyroid hormones in bone. *Endokrynologia Polska* 60: 380-388.
- Xu H, Watkins BA, Seifert MF. (1995). Vitamin E stimulates trabecular bone formation and alters epiphyseal cartilage morphometry. *Calcified tissue international* 57(4): 293-300.
- Xu Y, Lai X, Li Z, Zhang X, Luo Q. (2018). Effect of chronic heat stress on some physiological and immunological parameters in different breed of broilers. *Poultry Science* 97: 4073- 4082.
- Yan FF, Mohammed AA, Murugesan GR, Cheng HW. (2019). Effects of a dietary synbiotic inclusion on bone health in broilers subjected to cyclic heat stress episodes. *Poultry Science* 98: 1083-1089.
- Zeng H, Cao JJ, Combs GF. (2013). Selenium in bone health: Role in antioxidant protection and cell proliferation. *Nutrients* 5(1): 97-110.
- Zhang B, Guo X, Chen J, Zhao Y, Cong X, Jiang Z, Cao R, Cui K, Gao S, Tian W. (2014). Saikosaponin-D attenuates heat stress-induced oxidative damage in H1c-pk1 cells by increasing the expression of anti-oxidant enzymes and hsp72. *American Journal of Chinese Medicine* 42(5): 1261-1277.

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