

## The Effect of Perch Cooling and Perch Height on Some Bone Strength Parameters in Broilers Reared in Summer

M. Kenan TÜRKYILMAZ<sup>1,a,\*</sup>, Ahmet NAZLIGÜL<sup>1,b</sup>, Evrim DERELİ FİDAN<sup>1,c</sup>, Solmaz KARAARSLAN<sup>1,d</sup>, Mehmet KAYA<sup>1,e</sup>, Figen Sevil KİLİMCİ<sup>2,f</sup>

<sup>1</sup>Department of Animal Science, Faculty of Veterinary Medicine, Aydın Adnan Menderes University, Aydın, Turkey.

<sup>2</sup>Department of Anatomy, Faculty of Veterinary Medicine, Aydın Adnan Menderes University, Aydın, Turkey.

<sup>a</sup>ORCID: 0000-0002-7600-2390, <sup>b</sup>ORCID: 0000-0303-1476-4039, <sup>c</sup>ORCID: 0000-0002-9805-6162,

<sup>d</sup>ORCID: 0000-0002-6239-2439, <sup>e</sup>ORCID: 0000-0003-2377-4474, <sup>f</sup>ORCID: 0000-0002-2291-0545

Geliş Tarihi: 16.07.2020

Kabul Tarihi: 26.11.2020

**Abstract:** This study was carried out to investigate the effects of perch cooling and two different perch heights on physical (the weight, length, diameter, strength index, and weight-length index) and strength (strength, stiffness, elastic modulus, ultimate force) properties of tibiotarsus of broilers reared in summer conditions. A total of 450 one-d-old Ross-308 broiler chicks were used as the material. Chicks were allocated into 10 pens which has 15 chicks in each as to a 3x2x2 experimental design with three replication. Perch temperature was set to 10°C for cooled perches while perches were attached 7.5 and 15 cm off the floor. The position of both cooled and non-cooled perches were set to make animals pass over the perch for feeding and drinking. Results revealed that tibia weights in both 7.5 (19.39 g) and 15 cm (19.01 g) perch height groups were higher than the no-perch (17.24 g) group (P<0.001). Bones from the cooled perch group were found heavier (19.58 g-109.48 mm) and longer than the no-cooled (18.29 g-105.55 mm) group (P<0.001). As to ultimate force (F<sub>max</sub>), the greatest force (264.2 N) was required for bones in a group with 7.5 cm perch height (P<0.05). There were also similar significant differences in perch cooling and sex parameters while both perch application and perch cooling do not affect bone strength and elastic modulus.

**Key words:** Broiler, Bone, Cooling, Perch, Strength.

### Yaz Mevsiminde Yetiştirilen Etlik Piliçlerde Tünek Soğutma ve Tünek Yüksekliğinin Bazı Kemik Dayanıklılık Parametreleri Üzerine Etkisi

**Özet:** Bu çalışma, yaz koşullarında yetiştirilen etlik piliçlerde soğutulmuş tünek kullanımı ve tünek yüksekliğinin tibia kemiği fiziksel (ağırlık, uzunluk, çap, dayanıklılık ve ağırlık-uzunluk endeksi) ve dayanıklılık (dayanıklılık, katılık, esneme katsayısı ve kırılma gücü) özellikleri üzerine etkilerini incelemek üzere yapılmıştır. Bir günlük yaşta 450 Ross-308 civciv çalışmanın materyalini oluşturmuştur. Civcivler 10 ayrı bölmeye her bir grupta 15 civciv olacak şekilde 3x2x2 deneme düzeni çerçevesinde yerleştirilmiştir. Tünek sıcaklığı 10°C'ye ayarlanmış olup, tünekler tabandan 7.5 ve 15 cm yüksekte olacak şekilde konumlandırılmıştır. Soğutulmuş tünek ve yükseklik gruplarının tamamında tünekler hayvanların yem ve su gibi ihtiyaçlarını gidermek için üzerlerinden geçecekleri şekilde yerleştirilmiştir. Tibia ağırlığı hem 7.5 cm (19.39 g) hem 15 cm (19.01 g) tünek yüksekliklerinde tünek olmayan (17.24 g) gruptan daha yüksek çıkmıştır (P<0.001). Soğutmalı grup (19.58 g-109.48 mm) tibiaları soğutmasız (18.29 g-105.55 mm) olanlara göre hem daha ağır hem daha uzun olarak bulunmuştur (P<0,001). Kemiklerin kırılması için gereken güce gelince (F<sub>max</sub>); en yüksek güç değeri 264.2 N ile 7.5 cm tünek yüksekliği olan grupta gözlenmiştir (P<0.05). Benzeri önemlilik durumu tünek soğutma ve cinsiyet parametrelerinde bulunamamıştır. Aksine, hem tünek kullanımı hem tünek soğutmasının kemik dayanıklılığı ve dayanıklılık katsayısı üzerine herhangi bir etkisi olmadığı tespit edilmiştir.

**Anahtar Kelimeler:** Broyler, Dayanım, Kemik, Soğutma, Tünek.

### Introduction

Intense genetic selection and strict management practices led to a fast gain of muscle weight and tibial bones are loaded by heavy muscles and more prone to various disorders and even fractures (Charuta et al., 2011). But, the strength of leg bones is not only genetically determined but it also depends on the sex, age, health, nutrition, and environment. Hu et al. (2016) reported that thermally cooled perches reduced

thermoregulatory behaviors during acute heat stress, but did not affect their performance and physiological parameters under the ambient temperature imposed during the study. Kıyma et al. (2016) reported that perch use did not have any effect on growth performance but had a positive effect on footpad lesions. Enrichment of the environment such as perch usage can help broilers to overcome the negative effects of inactivity.

However, although broilers exhibited higher perching rates in the past (up to 27% of birds perching at 8 wk of age) (Hughes and Elson, 1977), later studies demonstrated much lower perching frequency (between 1.0 and 2.6%), (Su et al., 2000; Pettit-Riley and Estevez, 2001). It's thought that broilers failed to use perches in these studies because the effort required to jump up to the perch may have exceeded their ability or motivation to do so, especially as the increased weights of the birds exceeded what their legs could handle (Le Van et al., 2000). For this purpose, cooling the perches may generate the required motivation to broilers for perching.

In general, there is a need for a better understanding of some bone parameters in broilers under the influence of perch cooling and perch height in the same study. This study aimed to investigate the effects of perch cooling and two different perch heights on physical (the weight, length, diameter, strength index, and weight-length index) and strength (strength, stiffness, elastic modulus,  $F_{max}$ ) properties of tibiotarsus of broilers reared in summer.

## Material and Methods

### Material

Before experimenting, ethical clearance was obtained from the Institutional Animal Ethics Committee (ADU-HADYEK) of Adnan Menderes University, Aydin, Turkey (File No: 64583101/2016/121).

This study was carried out on 450 one-d-old Ross 308 broiler chicken in the Poultry Research Unit of Faculty of Veterinary Medicine, University of Aydin Adnan Menderes. A 3x2x2 (perch application x perch cooling x sex) factorial arrangement in a completely randomised design with three replication was used. All chicks were weighted and wing tagged at the arrival before their allocation into 10 different pens having 10 cm wood shavings. Animal density was set to 12 chicks/m<sup>2</sup> and ambient temperature and humidity were kept as to normal rearing conditions and recorded daily. All chicks were fed with commercial diets (3060 kcal/kg ME, 23% crude protein) and watered *ad libitum*. A standard lighting program (18L:6D) was applied after the first three days.

### Methods

A total of 200 right side tibia bones (20 from each pen) were used for physical bone properties. However, 5 bones were discarded from the measurements because of their problems. In terms of perch application, 8 steel perches (200 mm length and 30 mm diameter) were horizontally

attached 7.5 and 15 cm off the pen floor as to experimental design. The position of both cooled and non-cooled perches were set to make animals pass over the perch for feeding and drinking. Perch allowance per chicken was 15 cm. Out of eight perches, four perches were connected to the water cooler (Cihso 2000, 2.5 hp) which has capable of cool and circulate 5000 ml/min water at 10°C through the pipes. Water temperature in cooled perches was controlled twice daily.

**Three-point bending test:** On the 42<sup>nd</sup> day, 20 tibia bones from the right side of each chicken from each pen were cleaned of residual meat and other tissues. Each bone was weighed and measured for length and diameters with calipers. For the analysis of bone characteristics, these samples were wrapped with sterilised gauze dressing immersed in physiological saline solution and frozen at -20°C. Three-point bending test device (Zwick/Roell Z 0.5) in Adnan Menderes University Research Center (TARBIYOMER) was used to measure bone strength using advanced fulcra (ANSI/ASAE S459) for this purpose. Before the test, all bones were tawed. After the installation of the tibia into the device properly (total length/2), running speed was set to 10 mm/min and pre-load force to 2 N (Anonymous, 1993; Turner and Burr, 1993).

Bone deformation data till the breakage under the ultimate force were recorded. Force-deformation graphs for each bone were obtained from these data on these parameters: Ultimate force ( $F_{max}$ ): An amount of force at which the bone breaks (Newton-N). Stiffness: The slope of the elastic region of the force-deformation curve represents (Newton/meter-N/m). Bone strength: The maximum stress the bone can sustain is called the ultimate strength (GigaPascal-GPa) (Turner and Burr, 1993). Elastic modulus: Constant of proportionality between stress and strain (GPa). Moment of inertia: It is a measure of distribution of material around a given axis and is necessary to calculate the bending stress in a cross-section of the bone ( $lx m^4$ ) (Turner and Burr, 1993). The stiffness value for each bone was also calculated by using these graphs. The moment of inertia (I) which gives one to calculate the bending stress in the stressed cross-section of the tibia was calculated by measuring the inner and outer diameters of bone. Elastic modulus and strength values of bones were calculated by using force-deformation graphs and moment of inertia data (Anonymous, 1992; Harner and Wilson, 1986). About physical properties of bones; tibia weight, tibia length, diameter and strength index, and weight-length index were calculated as follows: Robusticity index: bone length/cubic root of bone weight. Weight-length

index: bone weight/bone length (mg/mm) (Karaarslan and Nazligul, 2015).

**Statistical analysis:** The data were analysed by the least-squares technique and the following mathematical model was designed to determine the effect of factors such as perch application, perch cooling, and sex affecting the traits under consideration:

$$Y_{ijkl} = \mu + a_i + b_j + c_k + e_{ijkl}$$

Where,  $Y_{ijkl}$  = Tibia strength, stiffness, elastic modulus,  $F_{max}$ , and moment of inertia, tibia weight, tibia length, tibia diameter, robusticity, and weight-length indexes.  $\mu$  = Overall mean,  $a_i$  = effect of perch application (no perch, 7.5 cm and 15 cm off the floor),  $b_j$  = effect of perch cooling (no cooled and cooled at 10°C),  $c_k$ : effect of sex (Male and female),  $e_{ijkl}$  = random error. Comparisons among subclass means were carried out by Duncan test available in SPSS (Version 22) (Duncan, 1955).

## Results

The results of the physical analysis of the tibiotarsus were given in Table 1. Tibia weight and length were measured higher in the group with 7.5 cm perch height than 15 cm and no perch groups ( $P < 0.001$ ). It should be noted from the table that perch cooling has significant effects on both tibia weight and length ( $P < 0.05$ ). As expected before, the weight and length of tibia for males were measured higher (21.53 and 109.72 mm, respectively) than female (16.12 and 104.52 mm, respectively) counterparts ( $P < 0.001$ ). Tibiotarsus diameters in the no-perch group were found lower (7.87 mm) than the other two groups (8.14 and 8.17 mm for groups with 7.5 and 15 cm perch height, respectively), ( $P < 0.05$ ). Different from perch height and sex, it's understood that perch cooling has no statistically significant effect on tibial diameter.

As to robusticity index calculations of these bones, there were no differences among perch height groups while this index in cooled perch (4.08) and female (4.15) groups was higher than non-cooled (4.03) and male (3.95) groups, respectively ( $P < 0.01$  and  $P < 0.001$ ). Weight-length index was calculated as the lowest in no perch group as parallel to weight and length values. Similar to tibia diameter, weight-length index has also no differences between cooled and non-cooled groups.

Ultimate force for the bones ( $F_{max}$ ) in groups with 7.5 and 15 cm perches was found higher (264.2 and 259.3 N, respectively) than no perch (226.7 N) group ( $P < 0.05$ ), (Table 2). It should be noted that perch cooling has a statistically significant effect on  $F_{max}$  ( $P < 0.05$ ) and there was a similar effect on sex. The effects and significances of stiffness and moment of inertia were found as similar to the

ultimate force for perch application, cooling, and sex. In contrast, it was determined that except sex, perch height and cooling have no effects on bone strength.

## Discussion

In general, the physical inspection of tibia bones revealed that broilers in perched groups (both 7.5 and 15 cm heights) have heavier, longer, and wider tibia bones in diameter than non-used counterparts while perch cooling has a significant effect on these parameters but diameter. In a study on the effect of lithium chloride on bone strength, Harvey et al. (2015) reported that average tibia length at the 42<sup>nd</sup> days was 120.0 mm for the control group. In another study, Gonzales et al. (2015) were found the tibia weight, length, and diameter as 13.14 g, 100.26 mm, and 7.49 mm for Arkansas random-bred chicken population, respectively. As parallel to this study, in a different study carried on the effects of dietary P restriction on tibia weight, length, and diameter revealed that birds given low P have shorter tibia (110.5 mm) than control counterparts (112.0 mm) (Moran and Todd, 1994). Kwiatkowska et al. (2018) also reported the range of tibia length and weight as 110.5 mm to 112.1 mm and 21.91 g to 23.45 g, respectively in a study on the effect of Fe-Glycinate chelate in diet for broiler chickens. But the same situation was not determined for tibia weight (18.0 g) and diameter (10.9 mm).

The results also showed that sex has an important effect on tibia weight, length, and diameter. The males had heavier, longer, and wider tibia bones in diameter compared to their female counterparts. As parallel to this study, Mabelebele et al. (2017) reported that tibia weight, length, and diameter were 36.91-25.58 g, 144.90-126.06 mm, and 8.79 -10.19 mm for male and female Ross-308 chickens at the age of 90 days, respectively. Charuta et al. (2011) who indicated that bone length was related to sex support these results.

The results indicated that perch cooling and sex have significant effects on the robusticity index. However, it's understood that weight-length index was only affected by perch height and sex. Although there was no statistical significance in the perch height group for robusticity index, it's seen that birds in no perch group have lower index than those in 7.5 and 15 cm perch height groups. A similar situation was valid for the weight-length index for perch cooling that birds in the cooled group have a higher index than those in the non-cooled group. The robusticity and weight-length index of tibia were found as 3.95-195.5 and 4.15-153.8 for males

**Table 1.** Some physical properties of tibiotarsus

Factors	n	Tibia weight (g)	Tibia length (mm)	Diameter (mm)	Robusticity index	Weight-length index
Exp. Mean( $\mu$ )		18.81	107.12	8.09	4.05	174.7
<b>Perch height</b>						
No perch	40	17.24 <sup>b</sup>	103.60 <sup>b</sup>	7.87 <sup>b</sup>	4.03	165.7 <sup>b</sup>
7.5 cm	78	19.39 <sup>a</sup>	108.31 <sup>a</sup>	8.14 <sup>a</sup>	4.05	178.1 <sup>a</sup>
15 cm	77	19.01 <sup>a</sup>	107.69 <sup>a</sup>	8.17 <sup>a</sup>	4.06	175.8 <sup>a</sup>
<b>Perch cooling</b>						
No cooling	116	18.29 <sup>b</sup>	105.55 <sup>b</sup>	8.11	4.03 <sup>b</sup>	172.4
10°C	79	19.58 <sup>a</sup>	109.48 <sup>a</sup>	8.07	4.08 <sup>a</sup>	178.0
<b>Sex</b>						
Male	98	21.53 <sup>a</sup>	109.72 <sup>a</sup>	8.60 <sup>a</sup>	3.95 <sup>b</sup>	195.5 <sup>a</sup>
Female	97	16.12 <sup>b</sup>	104.52 <sup>b</sup>	7.60 <sup>b</sup>	4.15 <sup>a</sup>	153.8 <sup>b</sup>
S <sub>x</sub>		0.162	0.320	0.049	0.008	1.17
P Values						
Perch height		0.001	0.003	0.024	0.976	0.005
Perch cooling		0.037	0.001	0.147	0.003	0.404
Sex		0.001	0.001	0.001	0.001	0.001

**Table 2.** Three point bending test results of tibiotarsus

Factors	n	F <sub>max</sub> (N)	Stiffness (N/m)	Bone Strength (GPa)	Elastic modulus (GPa)	I (m <sup>4</sup> )
Exp. Mean( $\mu$ )		254.8	77500.3	0.082	1.500	0.162
<b>Perch height</b>						
No perch	40	226.7 <sup>b</sup>	66742.8 <sup>b</sup>	0.088	1.643	0.128 <sup>b</sup>
7.5 cm	78	264.2 <sup>a</sup>	80717.2 <sup>a</sup>	0.082	1.484	0.171 <sup>a</sup>
15 cm	77	259.3 <sup>a</sup>	79662.2 <sup>a</sup>	0.081	1.445	0.170 <sup>a</sup>
<b>Perch cooling</b>						
No cooling	116	243.6 <sup>b</sup>	72955.8 <sup>b</sup>	0.084	1.578	0.150 <sup>b</sup>
10°C	79	271.5 <sup>a</sup>	84317.1 <sup>a</sup>	0.080	1.384	0.180 <sup>a</sup>
<b>Sex</b>						
Male	98	291.3 <sup>a</sup>	81484.8 <sup>a</sup>	0.078 <sup>b</sup>	1.180 <sup>b</sup>	0.200 <sup>a</sup>
Female	97	218.2 <sup>b</sup>	73515.8 <sup>b</sup>	0.087 <sup>a</sup>	1.821 <sup>a</sup>	0.124 <sup>b</sup>
S <sub>x</sub>		3.71	779.8	0.001	0.037	0.004
P Values						
Perch height		0.039	0.001	0.408	0.567	0.013
Perch cooling		0.020	0.001	0.404	0.055	0.024
Sex		0.001	0.001	0.007	0.001	0.001

and females, respectively. Mabalebele et al. (2017) reported the robusticity index and weight-length index as 4.35-254.84 and 4.28-207.81 for males and females, respectively. The difference between the two studies should be raised from the age of animals. As similar to the results of this study, Karaarslan and Nazligul (2015) reported that perch

usage has no significant effects on robusticity index (4.10 for perch, 4.11 for non-perch) and weight-length index (189.0 for perch, 186.89 for non-perch) while there was a statistically significant difference about sex for strength index (4.00 for males, 4.22 for females) and weight-length index (210.54 for males and 165.34 for females). It's well known that

the lower robusticity index means the stronger bone structure and the higher weight-length index means the denser bones. In terms of the physical properties of the tibia, it can be said that if the bone length continues to grow without the increase in bone width, this could make bones prone to breakage.

Bone tissue is composed of inorganic and organic matrices that provide support and mechanical strength. The inorganic matrix provides stiffness and compressional strength while the organic matrix (collagen) provides tensile strength and plastic property (Einhorne, 1996). As to three-point bending results, bones in the group with 7.5 cm perch height and cooled group have higher  $F_{max}$  (264.2 and 271.5 N, respectively) and stiffness (80717.2 and 84317.1 N/m). And sex has a significant effect on both ultimate force (291.3 and 218.2 N for males and females, respectively) and stiffness (81484.8 and 73515.8 N/m for males and females, respectively). Kwiatkowska et al. (2018) indicated that the ultimate force was ranged from 263.6 to 282.1 N. Harvey et al. (2015) reported the  $F_{max}$  and stiffness as 343.4 N and 253400 N/m for the control group. In that LiCl study, reduced stiffness could suggest a potential positive effect by reduced risk of fracture, and less energy was needed to bend the tibia bone of LiCl broilers as compared with controls implying a reduction in mineralization.

Higher moment of inertia may have arisen from the most commonly used perches were the ones cooled and with 7.5 cm height. Many studies reported that broilers have some problems with the transmission of the heat produced within their body beyond 25°C (Arad and Marder, 1982). Studies on the heat transfer ways of broilers showed that nearly 25% of total heat was transferred via their feet (Hilman and Scott, 1989). It's determined that when the ambient temperature rose, blood flow to feet also rose. In this respect, cooled perch would be very helpful for decreasing the body temperature using both perching and direct contact with wings or body. Also, Rath et al. (1999) reported that unlike cage layers, the birds reared in floor pens presumably had adequate movement and exercise, which are essential for the maintenance of bone strength and integrity. On the other hand, sex has also significant effects on  $F_{max}$ . The sex effect on  $F_{max}$  can be explained by the bigger diameters of the male bones. Apart from these, Martrenchar et al. (2000) reported that  $F_{max}$  was highly correlated to the bone weight ( $r=0.43$ ,  $P<0.01$ ) and to the live weight ( $r=0.45$ ,  $P<0.01$ ). However, it was unrelated to the presence of perches in the pen ( $F=299.7\pm 10.2$  N in birds that used perches frequently vs  $316.8\pm 9.1$  N in controls,  $P>0.05$ ). In another study about the effects of calcium and magnesium supplementation

on bone strength, there was no statistical difference among treatment groups and  $F_{max}$  was found as 247.90 N for the control group (Karasek et al., 2017).

In conclusion, it can be said that if it's height was set properly, perch application and perch cooling have great advantages for the motivation of fast-growing broilers which are prone to bone disorders.

## Acknowledgements

This manuscript was compiled from the project supported by Aydin Adnan Menderes University Scientific Research Projects Unit (Project Number: VTF-17005).

## References

- Anonymous, 1992: American Society of Agricultural and Biological Engineers, St. Joseph, Michigan, "Shear and Three-Point Bending Test of Animal Bone" ASAEANSI/ASAE.
- Arad Z, Marder J, 1982: Comparison of the productive performances of the Sinai Beduin fowl, the White Leghorn and their crossbreds: Study under natural desert conditions. *Br Poult Sci*, 23:333-338.
- Charuta A, Dzierzecka M, Majchrzak T, Czerwinski E, Cooper G, 2011: Computer-generated radiological imagery of the structure of the spongy substance in the postnatal development of the tibio-tarsal bones of the Peking domestic duck (*Anas platyrhynchos var. domestica*). *Poult Sci*, 90:830-835.
- Duncan DB, 1955: Multiple range test and multiple F-tests. *Biometrics*, 11: 1-42.
- Einhorne TA, 1996: Biomechanics of bone. In, Principles of Bone Biology. Raisz LG, Rodan GA, 25-27, Academic Press, USA.
- González-Cerón F, Rekaya R, Aggrey SE, 2015: Genetic analysis of bone quality traits and growth in a random mating broiler population. *Poult Sci*, 94: 883-889.
- Harner JP, Wilson JH, 1986: Testing techniques for determination of poultry bone strength. ASAE. *Am Soc Agr Biol Eng*, 29:642-644.
- Harvey BM, Eschbach M, Glynn EA, Kotha S, Darre M, Adams DJ, Ramanathan R, Mancini R, Govoni KE, 2015: Effect of daily lithium chloride administration on bone mass and strength in growing broiler chickens. *Poult Sci*, 94: 296-301.
- Hillman PE, Scott NR, 1989: Energy budget of the chicken foot. *Ther Biol*, 4:205-217.
- Hu JY, Hester PY, Makagon MM, Vezzoli G, Gates RS, Xiong YJ, Cheng HW, 2016: Cooled perch effects on performance and well-being traits in caged White Leghorn hens. *Poult Sci*, 95: 2737-2746.
- Hughes BO, Elson HA, 1977: The use of perches by broilers in floor pens. *Br Poult Sci*, 18: 715-722.
- Karaarslan S, Nazligul A, 2015: Effects of lighting, stocking density, and access to perches on leg health

- variables as welfare indicators in broiler chickens. *Livest Sci*, 218:31-36.
- Karasek F, Stenclova H, St'astnik O, Mrkwicova E, Pavlata L, Nedemova S, Zeman L, 2017: The effect of calcium and magnesium supplementation on performance and bone strength of broiler chickens. *Slov Food Sci*, 1:120-125.
- Kiyima Z, Küçükylmaz K, Orojpour A, 2016: Effects of perch availability on performance, carcass characteristics, and footpad lesions in broilers. *Arch Anim Breed*, 59: 19-25.
- Kwiatkowska K, Winiarska-Mieczan A, Kwiecień M, 2018: Effect of application of Fe-glycinate chelate in diet for broiler chickens in an amount covering 50 or 25% of the requirement on physical, morphometric and strength parameters of tibia bones. *Biol Trace Element Res*, 184: 483-490.
- Le Van NF, Estevez I, Stricklin WR, 2000: Use of horizontal and angled perches by broiler chickens. *Appl Anim Behav Sci*, 65:349-365.
- Mabelebele M, Norris D, Siwendu NA, Ng'ambi JW, Alabi OJ, Mbajjorgu CA, 2017: Bone morphometric parameters of the tibia and femur of indigenous and broiler chickens reared intensively. *Appl Eco Env Res*, 15:1387-1398.
- Martrenchar A, Huonnic D, Cotte JP, Boilletot E, Morisse JP, 2000: Influence of stocking density, artificial dusk and group size on the perching behaviour of broilers. *Br Poult Sci*, 41:125-130.
- Moran ET, Todd MC, 1994: Continuous submarginal phosphorus with broilers and the effect of preslaughter transportation: Carcass defects, further processing yields, and tibia-femur integrity. *Poult Sci*, 73: 1448-1457.
- Pettit-Riley RL, Estevez I, 2001: Effects of density on perching behaviour of broiler chickens. *Appl Anim Behav Sci*, 71:127-140, 2001.
- Su G, Sorensen P, Kestin SC, 2000: A note on the effects of perches and litter substrate on leg weakness in broiler chickens. *Poult Sci*, 79:1259-1263.
- Turner CH, Burr DB, 1993: Basic biomechanical measurements of bone: A tutorial. *Bone*, 14:595-608.
- \*Correspondence:** Prof. Dr. M.K. Turkyilmaz  
Department of Animal Science, Faculty of Veterinary Medicine, Aydin Adnan Menderes University, Isikli, Aydin-Turkey  
**e-mail:** mkturkyilmaz@adu.edu.tr