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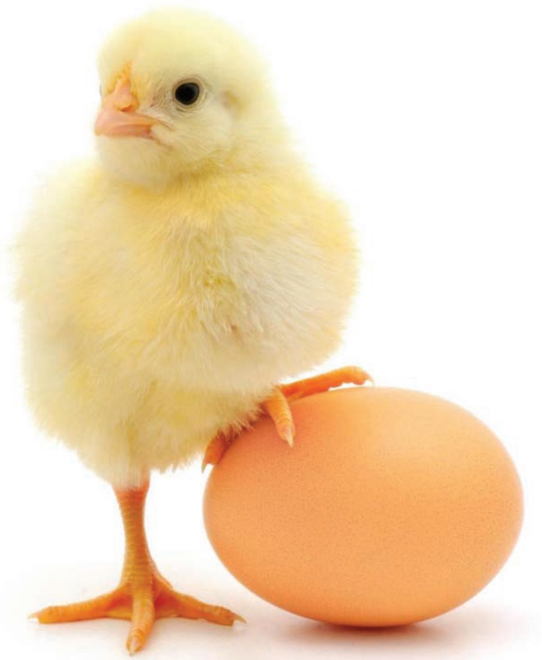

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Research Article
Free-Range Production System can Improve Bone Strength in Broilers

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Abstract

Background and Objective: Commercial broilers are raised for quick growth and enhanced feed efficiency. However, weight gain does not correlate with slower development of the skeletal system. The weight load causes skeletal problems in birds, forcing them to spend most of their time lying down and unable to express normal behavior. The goal of this research was to evaluate the effect of free-range production system (FRPS) on broilers' bone strength. **Materials and Methods:** The birds were distributed in a completely randomized design with 4 treatments (A, B, C and D) and 3 replications. Three birds were selected randomly from each replication, weighed and then euthanized fortnightly. Increase in skeletal load was estimated for each bird, right tibia de-fleshed, weighed and severed at the midpoint of its length. The inner and outer tibia midway diameters were measured, diameter ratios were (inner to outer) computed and expressed as percentages (IO). Tibia ash (%) and IO ratio were used to determine bone strength. **Results:** FRPS increased bone ash (%) and decreased skeletal load. Tibia ash (%) of the control treatment (D) and treatments A and B were significantly different ($p<0.05$). There was a positive correlation between tibia ash (%) and the IO ratio (Spearman's correlation of $r = 0.56$). **Conclusion:** The FRPS reduced the load on the bird's skeleton proportionally to its development stage, suggesting that FRPS increased the strength of bones, thus improved bird welfare.

Key words: Animal welfare, tibia bone ash, tibia bone diameter, musculoskeletal system, normal behaviour, skeletal load

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

In a free-range production system (FRPS), chickens have outdoor access, which promotes natural behavior and improves their welfare. In FRPS, chickens are raised in buildings that have outdoor pastures or ranges. This system meets consumer demand for ethically produced poultry products while improving the quality of life for the birds[1]. FRPS encourages exercise, improve locomotory and scavenging behavior[2]. Physical activity in broilers improves bone quality[3]. Broilers and pigs raised in FRPS exhibit superior locomotory behavior, including preening, pecking, dust bathing, walking and exploring, compared to animals in other production systems[4].

On the other hand, commercial broilers have been bred for rapid growth and enhanced feed efficiency since 1950s[5]. Broilers are genetically engineered to grow quickly and gain a lot of weight in a short time. However, as the birds mature, they spend long periods of their time lying down[6]. Confined broilers commonly show skeletal abnormalities such as soft and deformed bones, deformed limbs and a high incidence of bone breaking and death[7]. Skeleton of the broiler birds is crucial because it serves as the bird's support system[8].

Low-grade bones in broilers are primarily caused by low mineralization and a high number of empty spaces in the bone matrix, resulting in fragile bones prone to deformation and breakage[9]. Mineralization of bones strengthen them, allowing the skeleton to endure gravity and increased loading[10]. When birds suffer from lameness, they are unable to support their weight and they spend most of their time prostrate on the ground. Chronic exposure and improper litter handling have been linked to some of health issues, including limb deformities and atypical bones in broilers[11]. Bone is a dynamic living substance that undergoes bone cell remodeling during growth to respond to environmental factors such as changes in the bird's heaviness, physical activity, or mineral requirements[12]. Physical exercise can strengthen bones and reduce leg diseases[13]. Furthermore, in commercial broiler production systems, feeders and drinkers are placed near broilers to discourage broiler activity[14]. Bone strength is influenced by the volume of bone tissue, microstructure of the bone and the mineralization of the bone matrix[15]. Bone measurements such as bone tensile strength, bone density, bone calcium content[16] and bone ash[17] have been utilized as markers of bone health in poultry mineral nutrition. Bone with the highest mineral content is a good indicator of the mineralization of the skeleton[18]. Tibia bone ash (%) and the ratio of inner and outer diameters of the bones (IO) were used to determine if the free-range production system (FRPS) affected the bone strength of broiler birds. The objective of this study was to evaluate the effect of free-range production system on bone development in broiler birds.

MATERIALS AND METHODS

Study site: The study was carried out at the Pwani University poultry farm in Kenya, which is located at latitude 2° South, longitude 40° East, at an elevation of 16 m above sea level. The annual rainfall at the site is bimodal, ranging from 900-1100 mm, with an average temperature of 25-30°C[15].

Experimental design: A local hatchery (Kenchic Ltd) supplied 240 duly vaccinated cobb 500 broiler chicks for the experiment. For two weeks, the chicks were raised in total confinement on a deep litter floor system on a normal broiler diet.

The broiler chicks were distributed in a completely randomized design with four treatments (A, B, C and D) and 3 replications (20 birds each) at 2 weeks of age. The treatments were:

- **Treatment A:** Six weeks of FRPS
- **Treatment B:** Five weeks of FRPS
- **Treatment C:** Four weeks of FRPS
- **Treatment D:** Zero weeks of FRPS

All the birds received an initial vaccination against Newcastle disease at 14 days and a booster shot at 28 days of age. The birds in the FRPS were given access to grass and were free to roam from 7 A.M to 6 P.M and night was spent in confinement. The stocking density in the FRPS area was 20 square feet per bird. All birds received unlimited access to clean drinking water in all treatments.

Measurement of skeletal load: Three birds were selected at random from each replication (2, 4, 6 and 8 weeks of age) and weighed. Calculations were made on the weight gain at 4, 6 and 8 weeks of age (difference between the weights of the bird at weeks 4 and 2, 6 and 4 and 8 and 6 weeks, respectively).

This difference was recorded as an increase in skeletal load.

Tibia bone ash and diameters: Three birds were selected at random from each replication at 6, 7 and 8 weeks of age and were euthanized by stunning and cervical dislocation. The drumstick was detached from the stifle to the hock joint without causing damage to the tibia bone as stated by Rehman *et al.*[19]. Without boiling, the right tibia was extracted from the fresh carcass and the flesh was removed. Bone diameter was measured using the procedure described by Skinner and Waldroup[18] after slight modification. A hack saw was used to cut each tibia bone in half at the midpoint of its length. The midpoint of the tibia's length and diameter (both inner and outer) were measured using a Vernier caliper.

Then, the ratio of the inner to outer diameter was calculated and reported as a percentage. Afterward, two fragments of each tibia were placed in freezer bags and kept at -20°C for further analysis. The frozen bones were thawed, broken up into smaller pieces and dried in a dessicator (Jencons, Hertfordshire, United Kingdom) at room temperature for 24 hrs. To determine the amount of bone ash, approximately 3 g of the crushed bone was weighed into a porcelain crucible and initially ashed using a low-flame bunsen burner to remove the water and to char the sample thoroughly. The sample was finally ashed at 550°C in a muffle incinerator (Heraeus group, Hesse, Germany). To calculate the proportion of tibia ash, the weight of the tibia ash was divided by the dry weight of the tibia and multiplied by 100%[20].

Statistical analysis: Statistical analysis were performed using R programming language[21]. The Kruskal-Wallis test at $p<0.05$ was used to analyze the data as it was not normally distributed and Dunn's multiple comparison test was used to separate the means.

RESULTS

Weight of the broilers: All treatments showed a consistent rise in broiler weight across the weeks. (Fig. 1). However, the weight of the birds under FRPS were lower than those in confinement.

Skeletal load: Skeletal load of birds decreased as the duration on FRPS increased (Fig. 2). At the 4th week, the skeletal load was the lowest in treatment A (524 g), followed by treatment B (740 g). The skeletal load for treatments C (1223 g) and D (1229 g) was similar. At the 6th week, the increase in load in treatments A (150 g), B (299 g) and C (224 g) were much lower compared with the higher increase observed in treatment D (1394 g). At the 8th week, the skeletal load was lower for treatments A (143 g) and treatment B (92 g) but higher for treatment C (505 g). The skeletal load in treatment D dropped from 1394-435 g.

Tibia bone ash (%): For all treatments, tibia ash content (%) increased with age (Fig. 3). However, the increase was not the same for all treatments. The highest increase was observed in birds who spent the longest duration in the FRPS. At the 6th week, the tibia ash content was 34% (A), 25% (B), 14% (C) and 12% (D) for the different treatments. Similar trends were observed at the 7th week where the tibia ash was 33% (A), 27% (B), 17% (C) and 15 % (D) and at the 8th week, the tibia ash was 37% (A), 34% (B), 27% (C) and 17% (D) for the different treatments.

At the 6th week, differences ($p<0.05$) were recorded between treatments A and C, A and D a well as B and D (Table 1). Treatments A and B, B and C and C and D did not

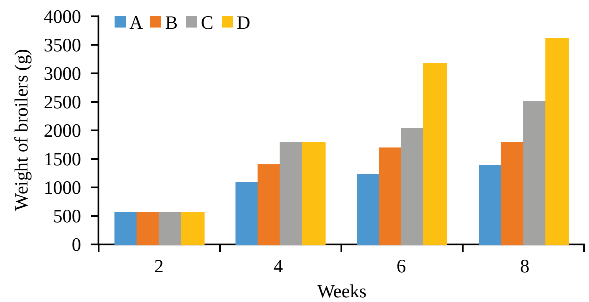

Fig. 1: The effects of FRPS on broiler weight at 2, 4, 6 and 8 weeks of age

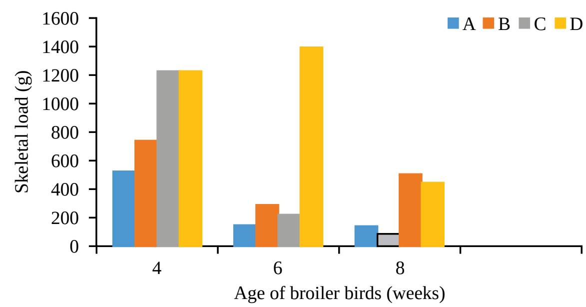

Fig. 2: Skeletal load of broiler birds at 4, 6 and 8 weeks of age

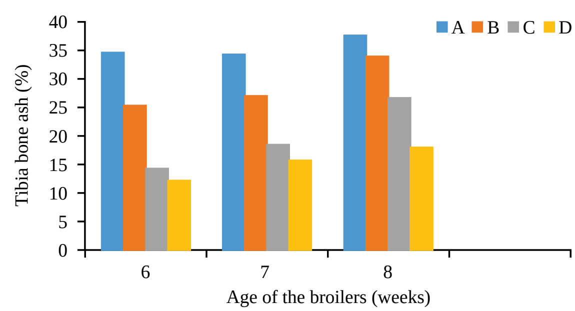

Fig. 3: Tibia bone ash (%) for different treatments at 6, 7 and 8 weeks of age of broilers

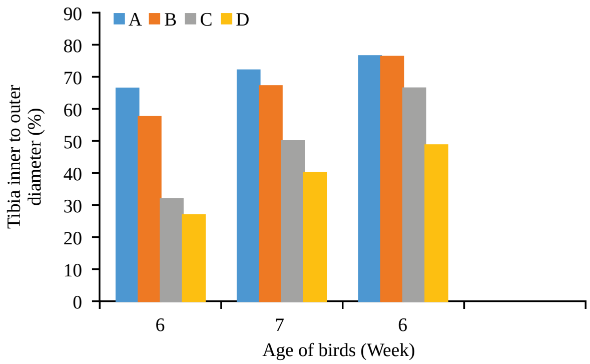

Fig. 4: Tibia IO for the different treatments at 6, 7 and 8 weeks of age

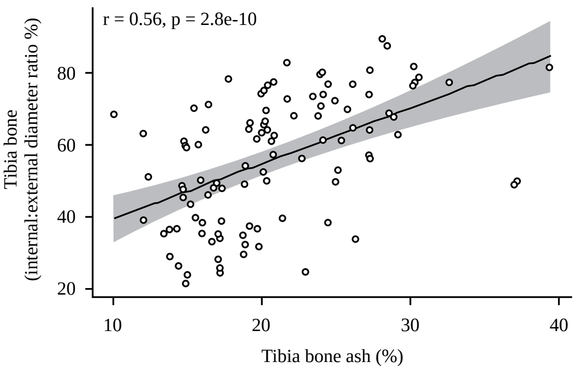

Fig. 5: Correlation between the inner diameter to outer diameter ratio (%) of the tibia bone (IO) and tibia bone ash content (%)

Table 1: Comparison of the percentages of tibia ash for the different treatments at 6, 7 and 8 weeks of age of broilers

Weeks	Comparison	Mean rank difference	p-value
6	A-B	7.67	0.736
	A-C	19.1	0.0007*
	A-D	24.6	0.0001*
	B-C	11.4	0.127
	B-D	16.9	0.004*
	C-D	5.44	0.999
7	A-B	6.56	0.999
	A-C	16.1	0.0071*
	A-D	25.1	0.0001*
	B-C	9.56	0.326
	B-D	18.6	0.0011*
	C-D	9	0.420
8	A-B	3	0.999
	A-C	14.4	0.0218*
	A-D	21	0.0001*
	B-C	11.4	0.127
	B-D	18	0.0017*
	C-D	6.56	0.999

*Indicate differences (p<0.05) between treatments

Table 2: Tibia inner to outer diameters as a percentage (IO) comparison for the different treatments at 6, 7 and 8 weeks of age of the broilers

Weeks	Comparisons	Mean rank difference	p-value
6	A-B	5.67	0.999
	A-C	18.4	0.0012*
	A-D	23.2	0.0001*
	B-C	12.8	0.0605
	B-D	17.6	0.0024*
	C-D	4.78	0.999
7	A-B	3.44	0.999
	A-C	16.4	0.0056*
	A-D	23.0	0.0001*
	B-C	13.0	0.0531
	B-D	19.6	0.0005*
	C-D	6.56	0.999
8	A-B	1.78	0.999
	A-C	12.2	0.0832
	A-D	22.7	0.0001*
	B-C	10.4	0.213
	B-D	20.9	0.0002*
	C-D	10.4	0.213

*Indicate the differences between the treatment percentage means (p<0.05)

differ (p>0.05). Similar results were observed at the 7th week. There were no significant differences (p>0.05) between treatments A and B, B and C as well as C and D. However, significant variations (p<0.05) were found between treatments A and C, A and D as well as B and D. There were significant differences (p<0.05) between treatments A and D and B and D at the 8th week but there were no differences (p>0.05) in the means of treatments A and B, A and C, B and C and C and D.

Tibia bone inner-to-outer diameter (%) (IO): There was a general increase in tibia bone inner to outer diameter (%) (IO) in all treatments over time (Fig. 4). However, the increase was not the same for all treatments. At the 6th week the IO was 65% (A), 55% (B), 32% (C) and 28% (D) for different treatments (Fig. 4). The same trends were observed at the 7th week where the IO was 71% (A), 67% (B), 50% (C) and 40% (D) and 8th week where IO was 77% (A), 76% (B), 65% (C) and 48% (D) for the different treatments.

At the 6th week, there were significant differences (p<0.05) between the treatments A and C, A and D and B and D. (Table 2). There were no significant variations (p>0.05) between the treatments A and B, B and C and C and D. At the 7th week, similar results were seen when differences (p<0.05) were recorded between treatments A and C, A and D and B and D. There were no significant variations between treatments A and B, B and C or C and D (p>0.05). There were differences (p<0.05) between treatments A and D and B and D at the 8th week but no differences (p>0.05) between treatments A and B, A and C, B and C and B and D. Figure 5 shows that there was a positive correlation between the IO and the bone ash (%).

The association between the IO and tibia ash (%) was positive (Spearman's r = 0.56 at p = 0.05).

DISCUSSION

Broiler weight and skeletal load: The current study aimed to investigate how FRPS affected body weight and bone strength and, ultimately, the welfare of broiler birds. The body weight of the broilers gradually increased with time but at varying rates depending on the treatment. However, FRPS reduced the broiler skeletal load; this result is in agreement with previous study[22], indicating that the skeletal load was lower in FRPS compared to the conventional broiler production system. While the skeletal loads of treatments A, B and C continued to reduce, treatment C recorded the lowest reduction. In week 4, birds in treatment C had already accumulated substantial skeletal load that impaired mobility, reducing their response to FRPS during the last 4 weeks. This result is congruent with a previous study conducted by Bokkers *et al.*[23], who found that a quick rise in broiler skeletal load impaired their capacity to behave according to any motivating factor, such as food seeking. Furthermore, around three weeks of age, broilers become progressively inactive due to a lack of incentives for energy-expending activities such as movement[24]. In other studies, it was hypothesized that early exposure to the FRPS during the early phase of broiler growth improved FRPS use in adult birds[25]. In other words, the earlier broiler birds are exposed to FRPS, the more they use it later in life.

Tibia bone ash (%): The tibia bone ash (%) was used to measure the strength of the bones due to ossification because previous studies have shown that mineralization of the bone matrix is one of the key determinants of the strength of bones[15]. In all treatments the tibia ash (%) increased with age, although at varying rates. This is in agreement with a previous study conducted by Skinner and Waldroup[18], who found that the percentage of tibia bone ash in broilers increased with time and, therefore, age of the birds. The rates of increase in the bone ash of the tibia were different due to changes in the skeletal load of the broilers. Increased physical activity improve bone quality by stimulating bone formation in chickens[3]. FRPS improved tibia bone ash (%) probably because birds have more opportunity to exercise. This is in line with Beattie *et al.*[4], who discovered that FRPS enhanced chicken (broilers and layers) and even pig activities like walking, investigating and dust bathing. The tibia ash (%) for treatments A and B, B and C and C and D did not differ at week 6 ($p>0.05$). This is likely because the birds were on FRPS for a short period of time. However, the tibia bone ash (%) between treatments A and C, A and D and B and D were different, ($p<0.05$) possibly because the FRPS exposure time was longer. Contrary studies have shown that bone development correlates with body weight to provide enough support for developing birds[17] but only true for slow-growing breeds of birds. The tibia bones of birds on FRPS grew rapidly, then slowed down by the end of 8th week. This is consistent with the findings of Barreiro *et al.*[26] who observed that skeletons develop more rapidly in the early stages of life than in the final phase. Other researchers found that physical activity decreases with time and lameness[6], reducing bone strength in later stages of the broilers' life.

In general, a rapid increase in muscle mass in broilers leads to skeletal problems, because bone strength cannot keep up with such a rapid muscle growth. Broilers spend almost all of their time recumbent on the ground on trash since they are unable to hold their weight. Most broiler problems are associated with prolonged exposure to litter[11]. Studies have indicated that reducing broiler growth rates, such as through feed limitation, can increase the strength of bones[9]. The FRPS, used in this study, is a form of feed restriction that reduced the growth rate of broilers and thus positively affected skeletal growth.

Tibia bone inner-to-outer diameter (%) (IO): Inner-to-outer diameter of the tibia bone (%) increased steadily with age in all treatments, although at different rates. The IO readings also showed that FRPS improved the IO, therefore, improving the bone health of the broilers. On the 6th week, there were significant variations ($p<0.05$) in the IO between treatments A and C, A and D and B and D, most likely due to the fact that birds were on FRPS for a long period of time. There were, nevertheless, no significant variations ($p>0.05$) in the means of treatments A and B, B and C and C and D.

The trends in tibia bone ash (%) were closely related to the trends in IO. The more the birds were in FRPS, the higher was their bone ash (%) and the higher was their IO. This was confirmed by a positive correlation between the IO and the bone ash (%). These two [tibia bone ash (%) and IO] can be used to assess the effect of FRPS on bone strength. Tibia bone ash (%) determination is time-consuming and is labour-intensive, both in terms of gathering the material and processing[27]. On the other hand, IO determination is faster with minimal labor and can be used as a tool for rapid results under field conditions. This correlation, implies that the IO can be used for quick assessment of bone strength in broiler birds.

CONCLUSION

Fast-growing broiler breeds are susceptible to bone anomalies and leg malformations due to their permeable skeletons. These foot disorders cause discomfort, restrict access to feed and water and lead to financial losses and welfare concerns. Free-range production systems (FRPS) help alleviate skeletal issues in broilers by reducing the skeletal load, improving bone strength and enhancing animal welfare. This knowledge can be used to develop innovative strategies for reducing bone disorders. Further research is needed to understand how different sexes of fast-growing broilers respond to treatments and address these issues in the chicken industry.

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